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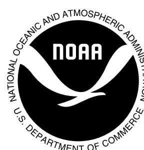

Contents

Acknowledgments v

Smoked Seafood Safety

Listeria monocytogenes: A Challenge for the Smoked Seafood Industry
Martin Wiedmann and Ken Gall 1

Clostridium botulinum Concerns
Mary Losikoff 5

Staphylococcus aureus Concerns in Smoked Fish
Brian H. Himelbloom 9

A Review: Processing Parameters Needed to Control Pathogens in Cold Smoked Fish
Michael Jahncke 15

Bacteriophage Control of Pathogens in Foods
Jenny Scott and Yuhann Chen 23

Pellicle Formation and Inactivation of *Listeria* and
Staphylococcus Species in Hot-Smoking of Salmon
Brian H. Himelbloom, Chuck Crapo, Thombathu S. Shetty, and Carey Vorholt 27

Regulatory Standards

International Regulatory Requirements for Smoked
Fish: Europe and Codex Alimentarius
Timothy Hansen 35

HACCP Plan
Jim Vik 39

Compliance with FDA's HACCP Regulation
Christopher E. Rezendes 43

Process Establishment or Validation for Refrigerated, Vacuum-Packaged
Hot-Smoked Fish, and HACCP Monitoring Considerations
Joe Frazier 47

Common Mistakes in HACCP for Hot Smoked Salmon
Liz Brown 53

Production and Packaging

Wood Smoke Components and Functional Properties
Romeo T. Toledo 55

Introduction to Smoke Condensates
Jeffrey J. Rozum 63

Food Engineering Applications in Seafood Processing
Subramaniam Sathivel 65

Development and Characterization of Vacuum Packaged Alaska Pink Salmon (*Oncorhynchus gorbuscha*) Jerkies Made with Marinades
A.C.M. Oliveira, C.A. Crapo, B.H. Himelbloom, A. Morey, and A. Ambardekar 67

Using Sustainable Packaging Technologies to Respond to Consumer, Retailer, and Seafood Industry Needs
Barbara Blakistone and Claire Koelsch Sand 75

Use of Antimicrobial-Coated Plastic Films to Control *Listeria monocytogenes* on Cold-Smoked Salmon
Hudaa Neetoo, Mu Ye, and Haiqiang Chen 81

Business and Marketing

Globalization and Aquaculture: Challenges, Opportunities, and Questions for the Smoked Seafood Industry
Gunnar Knapp 91

Fishing for Answers? Consult the Internet!
Pamela D. Tom 95

Planning, Constructing, and Managing a Community Freezing and Cold Storage Facility: A Case Study
Sunny Rice and Eric Philips 99

RFID: How It Will Transform Packaging, Distribution, and Handling of Alaska Seafood
Stephen T. Grabacki, Mike Ronchetti, Thane Humphrey, and Oliver Hedgepeth 101

USDA Rural Development: Value Added Producer Grant (VAPG) Program
Karen Dearlove 107

Innovations in Alaska Smoked Seafood as Showcased through the Alaska Symphony of Seafood
Bob Pawlowski and Jim Browning 111

Participants 115

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Listeria monocytogenes: A Challenge for the Smoked Seafood Industry

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Research summary

Listeria monocytogenes causes an estimated 2,000 to 2,500 cases of illness per year in the United States. Since about 20 to 30% of listeriosis patients die, this is one of the most severe food-borne diseases. Susceptible individuals (e.g., immunocompromised people and pregnant women) can experience severe disease, including abortions and brain infections, when exposed to high numbers of this bacterium. *L. monocytogenes* and the disease caused by it have a number of characteristics that complicate efforts to reduce food-borne listeriosis infections.

The almost ubiquitous presence of the organism, including in urban and natural environments, farms, and processing plants, provides a high risk for introduction into foods at any point of the farm-to-table food continuum. L. monocytogenes' ability to multiply in foods at refrigeration temperatures allow this organism to reach high numbers, particularly if foods that otherwise allow its growth (e.g., by having suitable pH and water activity) are stored for prolonged times at refrigeration temperatures. If these foods are not heated before consumption (as is generally the case for ready-to-eat foods) consumers can then be exposed to high *L. monocytogenes* numbers. The long incubation period for listeriosis (7 to 60 days) makes it difficult to identify the sources of infection and therefore complicates efforts to identify and eliminate sources of human infections.

We have used molecular subtyping methods (as reviewed by Wiedmann 2002) to explore the sources and spread of *L. monocytogenes* in a variety of environments (e.g., Kabuki et al. 2004) including in smoked seafood processing plants (Hoffman et al. 2003, Lappi et al. 2004). While we found a variety of different transient *L. monocytogenes* subtypes in many plants, a specific *L. monocytogenes* subtype or subtypes that persist in the processing environment was also identified in most processing plants. These persistent subtypes appear to be the major cause of finished product contamination.

While molecular subtyping helped identify likely niches for persistent subtypes, control and eliminations of these persistent *L. monocytogenes* appears to represent a major challenge in all sectors of the food industry. In addition, we identified considerable variation in *Listeria* contamination patterns in a smoked seafood plant within a given day and within a given shift. This further complicates *Listeria* control efforts, and indicates the importance of random events in *Listeria* transmission (Hu et al. 2006). Despite these considerable challenges, long-term efforts to control *L. monocytogenes* in one plant led to a reduction of *L. monocytogenes* prevalence to approximately one-tenth, indicating that improved control of this pathogen in the seafood industry is possible.

Control strategies for smoked seafood processors

Listeria monocytogenes can survive for long periods of time in the processing plant environment. It may be introduced into processing plants through a variety of routes, including raw materials, employees, and equipment. *Listeria* species tend to form a biofilm, which enhances its survival when resident populations become established in niches in the plant. General cleaning and sanitizing procedures do not easily eliminate these resident populations.

Implementing an effective *Listeria* control program is a long-term commitment. Based on our current understanding, at least five key elements need to be included in an effective *L. monocytogenes* control program for ready-to-eat seafood products like smoked fish. These five elements include (1) *Listeria* specific good manufacturing practices (GMPs) and sanitation procedures, (2) employee training, (3) environmental microbiological monitoring and testing, (4) raw material controls, and (5) controls to minimize growth in the finished products.

The Smoked Seafood Working Group (SSWG), a collaboration of two national industry trade organizations, the U.S. National Fisheries Institute and National Food Processors Association, smoked seafood processors, and academia, developed guidelines to minimize *Listeria monocytogenes* contamination of finished products in smoked seafood operations. These guidelines have been adopted by reference in the Association of Food and Drug Officials' Cured, Salted and Smoked Fish Good Manufacturing Establishments GMPs (AFDO 2004). The SSWG (2002) *Listeria* Control Manual can be downloaded from the following Cornell University Web site: http://www.foodscience.cornell.edu/

cals/foodsci/research/labs/wiedmann/listeria-employee-training-program.cfm. The control strategies developed by the SSWG for *Listeria*-specific GMPs and sanitation controls, employee training, microbiological testing of the plant environment, raw materials and finished products, and antimicrobial treatments for raw material and finished products have been published in a series of four papers in the peer reviewed journal of the International Association for Food Protection, *Food Protection Trends*.

An Institute of Food Technologists' expert panel stated that reduction of *L. monocytogenes* in the processing plant was directly dependent on adherence to good hygienic practices (GHPs) and GMPs (IFT 2001). Targeted good manufacturing practices and sanitation procedures to minimize *Listeria* contamination of smoked seafood products are summarized in Gall et al. 2004. These procedures include steps to prevent cross contamination caused by improper design and layout of processing operations, the movement of people and equipment in the plant, inadequate employee hygiene, and poor food handling practices. Cleaning and sanitation procedures for equipment and the processing plant environment that are designed to specifically target *Listeria* contamination are also described.

Training plant personnel is another key element of the complete *Listeria* control program identified by the SSWG. Examples of targeted training programs for all plant employees, those employees who work with exposed finished products, and employees who conduct cleaning and sanitation procedures are described in Hicks et al. 2004. Three different PowerPoint training programs that are designed to train employees in each of these groups in the plant environment can be downloaded free of charge at the following Cornell University Web site: http://www.foodscience.cornell.edu/cals/foodsci/research/labs/wiedmann/listeria-employee-training-program.cfm.

Another element of a complete *Listeria* control program that smoked seafood processors may need to consider is treatment with antimicrobial agents to reduce the amount of *Listeria* on raw seafood products or treatments to inhibit its growth on finished packaged smoked seafood products. A scientific and practical review of the various antimicrobial treatment options that may be available to smoked seafood processors is provided in Jahncke et al. 2004.

Environmental testing for *Listeria* can be used to help identify problem areas or locate contamination sources in the processing plant, and to confirm that problem solving procedures have been effective. It may also be necessary in some circumstances to test raw materials and finished products. Guidelines for *Listeria* testing of environmental and raw and finished product samples in a smoked seafood processing environment are provided in Scott et al. 2005.

Controlling *Listeria monocytogenes* in both cold and hot smoked seafood products can be a significant challenge. For cold smoked products, the heat applied during the smoking process is not sufficient to kill *Listeria*, so controls must be in place to minimize *Listeria* contamination of raw materials and at all steps of the process. Hot smoked products are subjected to a heat treatment that is lethal to *Listeria*, but these products are susceptible to contamination from the plant environment after smoking. Thus control efforts should focus on preventing contamination after the hot smoking step.

Each firm needs to evaluate their unique situation and implement targeted control strategies for their plant environment and the products that they produce. The resources listed in the references below can be used by smoked seafood processors to help them build an appropriate science-based *Listeria* control program.

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Clostridium botulinum Concerns

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Clostridium botulinum is a group of culturally distinct organisms that produce similar toxins. It is an anaerobic spore-forming bacterium and the toxin produced is a neurotoxin that is considered one of the most potent naturally occurring toxins known. The vegetative cells and the toxin are susceptible to heat, while the spores can be heat resistant and can survive adverse conditions. The bacteria are widely distributed in nature in soils; sediments of streams, lakes, and coastal waters; the intestinal tracts of fish and mammals; and the gills and viscera of crabs and other shellfish.

There are seven types of *C. botulinum*—A, B, C, D, E, F, and G—while only four of those, A, B, E, and F, cause human botulism. There is a low incidence of disease but the mortality rate is high if the symptoms are not immediately treated. The onset of symptoms is from 18 to 36 hours and early signs are weakness and vertigo, leading to progressive paralysis. Some victims experience abdominal distention and constipation. Victims will experience double vision, and difficulty in speaking and swallowing and breathing, eventually leading to inhibited respiration and death from asphyxia. Cases may be under-diagnosed or missed entirely because the disease can occur as a single case and the symptoms are similar to other illnesses, such as stroke. Treatment includes early administration of botulinal antitoxin and mechanical ventilation. Early administration is important because the antitoxin works against toxin that has not bound to the nerve ending. Two to eight weeks of ventilation support is common.

C. botulinum is also differentiated by its ability to degrade proteins. The proteolytic strains (A, B, F) will degrade proteins, leaving visual signs of growth. The limit for growth (temperature below which it will not grow) for the proteolytic strains is 10°C (50°F). The non-proteolytic strains (B, E, F) do not degrade protein so the product may be toxic without signs of growth. The limit for growth for the non-proteolytic strains is 3.3°C (38°F) which is below recommended refrigeration temperatures. The strains most commonly associated with seafood products and the marine environment are non-proteolytic with type E of particular concern.

The organisms are ubiquitous in the environment. Non-proteolytic strains of *C. botulinum* were found in 42.6% of Pacific rockfish and salmon sampled in a 1990 study. The bacteria were primarily found in the guts and gills so evisceration is a key to control. Table 1 shows examples of levels of spores found in several species of fish.

Table 1. *Clostridium botulinum* spore levels in fish.

Fish	Strain	Spore level
Pacific rockfish (red snapper)	A	9-240/100 g
Salmon	A	4-120/100 g
Herring	E	100/100 g
Farm raised trout from Sweden	E	530/100 g

(Baker et al. 1990)

The United States has relatively low incidences of food-borne botulism cases, with CDC reporting 1,087 cases in 444 outbreaks recorded between 1950 and 1996. An outbreak is defined as one or more cases of botulism where a food source is implicated. There is an average of 2.5 cases per outbreak. Of these 444 outbreaks, 37.6% were type A, 13.7% were type B, 15.1% were type E, 0.7% were type F, and 32.9% were unknown. Five states (California, Washington, Colorado, Oregon, Alaska) are responsible for more than half (53.8%) the outbreaks. When you isolate data from 1990-1996 outbreaks, type A was responsible for 44.6%, type E for 35.7%, and type B for 12%. This may be due to better isolation techniques. Earlier in the century, there was a 60% fatality rate. Between 1950 and 1996, the fatality rate dropped to 15.5%, which could be due to improvements in respiratory intensive care and the prompt administration of antitoxin. Alaska was responsible for 16.2% of outbreaks nationwide between 1950 and 1996, largely due to improper preparation and storage of Alaska Native foods. Fifty-six (83.6%) of the nationwide type E outbreaks took place in Alaska.

Selected cases in the United States

Early in the 1960s, there were three episodes of botulism from vacuum packaged smoked fish from the Great Lakes, which resulted in 21 cases with nine deaths. In 1987, consumption of Kapchunka, an uneviscerated, air-dried whole whitefish caused eight cases of botulism in New York and Israel with one death. In Hawaii, in 1990, three people became ill after consuming grilled palani. The investigation showed that apparently toxin was formed in the raw fish after it was temperature abused in the market; the heat applied during cooking did not inactivate the botulinal toxin.

Selected case study

This incident in Europe provides an overview of a relatively recent case of botulism in a commercially prepared smoked

fish product. The authors of the study referred to vacuum packaged hot-smoked fish as "one of the most important botulism food vehicles processed on an industrial scale." In 1997, a family of three ate from a package of vacuum packed, hot-smoked whitefish. The wife, who consumed two-thirds of the fish, became ill in 7.5 hours. In 13.5 hours the husband experienced symptoms, while the two-year-old grandchild was unaffected. The couple said the fish "tasted good." The remains of the package were positive for type E *C. botulinum* and toxin. Eleven other samples from same lot tested negative. This suggests that contamination can be spotty and food processors must take care to ensure that smoked fish in each package is properly brined, smoked, and heated. The whitefish had been processed five days prior to consumption. The process included thawing the raw product in cold water, brining for ten hours at 5°C, drying at 40°C for 30 minutes, and smoking in four steps between 60° and 75°C for about two hours. The resulting product had a water phase salt level of 1.8% and was sold in Germany and Finland. There were no documented product temperatures from the processor or the distribution chain, but when measured later, storage temperatures ranged from 2° to 5°C. Conclusions from the investigation indicated low water phase salt (wps) with no obvious temperature abuse. There was some indication that the processing, storage, and transport information may not have been correct and there was a possibility of preformed toxin. The lack of specific information demonstrates the value of accurate HACCP records.

The hazard of *C. botulinum* in seafood products is regulated through the Seafood HACCP Regulation (21 CFR Part 123), and guidance to the industry is provided in the Fish and Fishery Products Hazards and Controls Guide, 3rd edition, 2001. A new edition is expected to be published in 2008. The guide addresses the *C. botulinum* hazards associated with the use of reduced oxygen packaging, including vacuum packaging, modified atmosphere packaging, hermetically sealed containers (e.g., double seamed cans and glass jars with lids), deep containers from which air is expressed, and products packed in oil. FDA's concern is that the use of reduced oxygen packaging (ROP) may extend shelf life by inhibiting the growth of aerobic spoilage organisms, and the anaerobic conditions created may allow the growth and toxin production by non-proteolytic strains of *C. botulinum* at refrigeration temperatures without visible signs of spoilage.

Controls to prevent the growth of *C. botulinum* include freezing (where labeling is a key), heating (sufficiently to inactivate spores), acidifying, reducing the water activity through drying, adding chemicals such as salt, refrigerating the food, or some combination of these factors. The controls for complete inhibition differ between proteolytic and non-proteolytic strains as shown in Table 2.

Table 2. Controls for complete inhibition of *Clostridium botulinum* proteolytic and non-proteolytic strains.

	Proteolytic strains A, B, F	Non-proteolytic strains B, E, F
pH	<4.6	<5.0
a_w	<0.94	<0.97
NaCl	10% wps	5% wps
Min temp.	10°C (50°F)	3.3°C (38°F)
Spore heat resistance	High: D value 121°C = 0.23 min	Low: D value 82.2°C = 1.2 min

Heat can be used to eliminate non-proteolytic strains of *C. botulinum* when it is applied to product in the final package (pasteurization). The product is then refrigerated at 4.4°C (40°F) to prevent the outgrowth of other pathogens and the proteolytic strains of *C. botulinum*. Products that have been successfully treated in this manner include surimi analogs, pasteurized crabmeat, and cooked shrimp.

Non-proteolytic strains are completely inhibited in fish salted to a 5.0% wps, acidified with a pH of 5.0 or less, or water activity controlled below 0.97. In each case the product must be refrigerated at 40°F (4.4°C) to inhibit other pathogens that may be present, including proteolytic strains of *C. botulinum*. Chemical inhibitors can also be used at lower levels against non-proteolytic *C. botulinum* when combined with other treatments, as in the case in smoked fish, which incorporates smoke and temperature with 3.5% salt.

Water phase salt is the amount of salt in the product relative to the product moisture and is found using the following calculation.

$$\%\ \text{water phase salt} = \frac{\%\ \text{salt} \times 100}{\%\ \text{salt} + \%\ \text{moisture}}$$

In hot smoked fish, controls include a combination of salt, smoke, and heat to achieve a wps of 3.5%. The temperature needed in a hot smoke process is 145°F for 30 minutes. The cooking oven needs to be tested to find the cold spot, and three probes are recommended to measure the smoking process in the coldest spot and the thickest part of the fish. After processing, the protein in hot smoked fish is coagulated.

Cold smoked fish needs 3.5% wps, and the smoker temperature should not exceed 90°F. The smoking temperature must be low enough to prevent coagulation of the protein and elimination of the spoilage flora. The resulting product has a raw appearance.

A processor needs to conduct scientific studies to determine the factors needed to consistently achieve the appropriate water phase salt levels in a given product. The studies should address factors such as the minimum brining time, the minimum salt concentration in the brine, the minimum ratio of brine to fish, the maximum fish thickness or size, the influence of drying and smoking, and restricting a load to a single species.

It is key to the safety of both cold and hot smoked fish products for the processor, to manage the amount of time the products are exposed to temperatures favorable to *C. botulinum* growth and toxin formation during processing, during

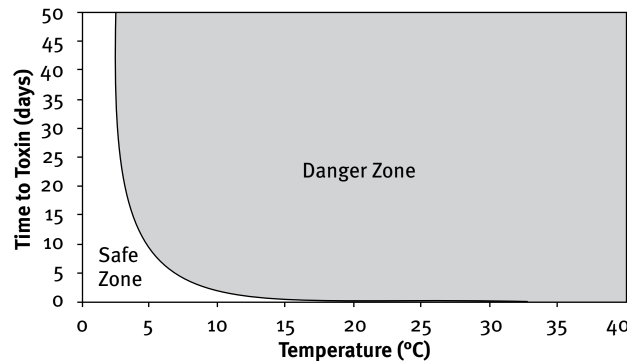

Figure 1. Skinner and Larkin curve (Skinner and Larkin 1998).

finished product storage, and at receipt by the secondary processor. The Skinner and Larkin curve (Fig. 1) shows time to toxin formation at various incubation temperatures.

If refrigeration is being used as the sole barrier to *C. botulinum* growth, the critical limit for the packaged product is storage at or below 38°F (3.3°C) and the temperature needs to be monitored. One method would be to use a time temperature integrator or indicator on each package. A time temperature integrator (TTI) would demonstrate through a color change or other visual means that the product has been exposed to time and temperature conditions that may allow the toxin formation by *C. botulinum*. An indicator generally shows that a given temperature has been exceeded. The processor is responsible for choosing a TTI that has been validated by the manufacturer. This validation should demonstrate that the TTI is functional and fit for its intended purpose taking into account:

- Closely matches the Skinner and Larkin curve.
- Level of confidence, false negative rate.
- Standard deviation of the lot.
- Limitations of the TTI—prevent exposure to humidity, heat, storage, etc.
- Descriptive insert—applicability (uses), interpretation criteria, shelf life of the TTIs, environmental factors, etc.

Factors defined by the validation, e.g., a need to maintain a given TTI frozen prior to application and other limitations of the TTI, would become part of the processor's HACCP plan. Other examples of critical control points (CCPs) may include the application and the activation of the TTI. Records should include challenges to verify that each lot of TTIs is functioning, as well as records of receipt and shelf life of TTIs. The processor must know the limitations of the TTI and address those limitations in their HACCP plan.

Packaging specifications may state that a given packaging film is oxygen permeable. There are degrees of oxygen permeability, and it is affected by various factors including temperature and film thickness. Most specifications list an oxygen transmission rate (OTR). An example of an oxygen impermeable package would be one having an OTR of 100 cc per m² per 24 hours at 24°C. An example of an oxygen permeable package would be one having an oxygen transmission rate (OTR) of 10,000 cc per m² per 24 hours at 24°C. The Hazards Guide suggests that packaging material with an OTR greater than 10,000 cc per m² per 24 hours at 24°C should provide sufficient oxygen exchange to allow aerobic spoilage organisms to grow and spoil the product prior to toxin formation. Industry members have the option of providing data to establish a different OTR for products. It should be noted that OTR does not compensate for products packed in oil, which will restrict oxygen exchange.

Nitrites may only be used in salmon, sable, shad, chubs, and tuna according to FDA Compliance Policy Guide sections (21 CFR 172.175). When nitrites are used, the salt level can be reduced, i.e., a level of 3.0% wps in combination with not less than 100 ppm nitrite is required.

In summary, for those of you considering smoking fish for the first time, I want to emphasize that *C. botulinum* type E has been associated with botulism cases in Alaska. While associated with Native Alaskan foods, it demonstrates that the organism is present and must be addressed. Salt levels, smoking, drying times and temperatures, and storage temperatures are critical to the safety of the smoked fish. HACCP and its recordkeeping requirements are key. Evisceration is essential because that is where the spores are.

Finally, a word of caution to smokers who use mail-order for shipment of product. Often product will arrive at its destination at ambient temperature. A single small gel pack is seldom effective. Several adequate gel packs or dry ice or styrofoam containers may be necessary. The processor needs to check to ensure that the product arrives at its destination at the appropriate temperatures even in the warmest climates.

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Staphylococcus aureus Concerns in Smoked Fish

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Abstract

The well-known food-borne pathogen, *Staphylococcus aureus*, rarely has been implicated in cases originating from consumption of smoked seafoods. This bacterium may be contributed through human handling of the raw seafood and products. Nevertheless, adequate precautions can prevent *S. aureus* contamination, growth and enterotoxin production from occurring in smoked fish products. A literature review was conducted to illustrate the incidence of *S. aureus* relative to contamination of smoked seafoods and the pathogen's contribution to the likelihood of seafood-borne illnesses in consumers.

Human pathogens in seafoods

There are only a few bacteria that are associated with causing food-borne illnesses from consuming pathogen-containing seafood. *Clostridium botulinum*, *Listeria monocytogenes*, three *Vibrio* species, and *Salmonella* species are the most common types. *Staphylococcus aureus* remains a questionable or low degree of concern since this bacterium is considered nonindigenous to marine fish and shellfish (Kvenberg 1991, Huss et al. 1995, Sanjeev and Surendan 1996, Huss et al. 2000). Furthermore, *Listeria monocytogenes*, *C. botulinum* type E, biogenic amines, and parasites were the only concerns in a report on the safety of cold-smoked fish (IFT 2001).

Staphylococcus aureus, enterotoxins, and epidemiology

Over 30 species of *Staphylococcus* are known and are characterized as Gram-positive cocci, non-motile, non-sporeforming, facultative anaerobes (Baird-Parker 2000). The main reservoirs for *S. aureus* are skin and mucous membranes of warm-blooded animals including humans (Smith et al. 1983, Baird-Parker 2000). Four biotypes (A to D) have been differentiated however all coagulate animal blood plasma, produce a heat-resistant nuclease and type A hemolysin is specific for humans (Baird-Parker 2000). Enterotoxin types A to H synthesized by *S. aureus* are heat-tolerant, freeze-tolerant, and protease-resistant proteins (Smith et al. 1983, Baird-Parker 2000). Recent advances in genomic analysis have expanded the list to 21 staphylococcal enterotoxic antigens (Seo and Bohach 2007). The minimum amount of enterotoxin to cause outbreaks is 1 μg which results from 10^6 cells per g (Smith et al. 1983, Baird-Parker 2000) although lower amounts (0.001-0.1 μg per g of food) may cause illness in susceptible people (Smith et al. 1983, Seo and Bohach 2007). Symptoms of enterotoxicity include nausea, vomiting, abdominal pain and diarrhea but no fever within 1-8 hours (Baird-Parker 2000).

Epidemiological history of *S. aureus* shows staphylococcal intoxications from a variety of fish products in ten countries as early as the 1930s and 1940s (Shewan 1962) and caused by fish salads and shellfish salads in the 1950s, 1960s, and 1970s (Masi et al. 1959; La Chapelle et al. 1966; Minor and Marth 1972; Bryan 1973,1980). During the 1970s in the United States, *S. aureus* was the most frequent food-borne illness (Baird-Parker 2000). According to the Centers for Disease Control and Prevention, *S. aureus* accounts for 185,000 food-borne illnesses per year. It ranks fifth for all bacterial pathogens and lags several viruses and parasites (Mead et al. 1999). Cooked meats, poultry, seafood (crabmeat, shrimp), and cream-filled bakery products are the most common foods implicated in *S. aureus* intoxications (Baird-Parker 2000). Potentially hazardous foods may be as common as one-third of cooked seafoods prepared in Egyptian food establishments (Saddik et al. 1985). *Staphylococcus aureus* strains, capable of producing enterotoxins A, C, and/or D primarily, could be isolated from the palms and throats of half of the fish processing workers from six Indian factories (Sanjeev et al. 1987). Food workers in restaurants worldwide caused 53 outbreaks, which contributed to over 6,400 cases of *S. aureus* food-borne illness; however, no seafood products were implicated (Greig et al. 2007).

Smoked fish and *S. aureus*

Only one documented report showed *S. aureus* to be the causative agent in smoked seafood. Hot-smoked (100°C for 30-60 min) South African snoek, *Thyrsites atun*, caused illnesses in two people (Simmonds et al. 1973, Prior et al. 1977). The problem was attributed to post-processing contamination at the wholesale smokehouse and temperature abuse during storage (Lamprecht and de Chaville 1973, Prior et al. 1977, Simmonds et al. 1974). The water activity (a_w) of the product was 0.93, sufficient to allow growth of at least 2×10^5 *S. aureus* cells per g (Prior et al. 1977) and it is theorized that the enterotoxin dosage was enough to cause illness.

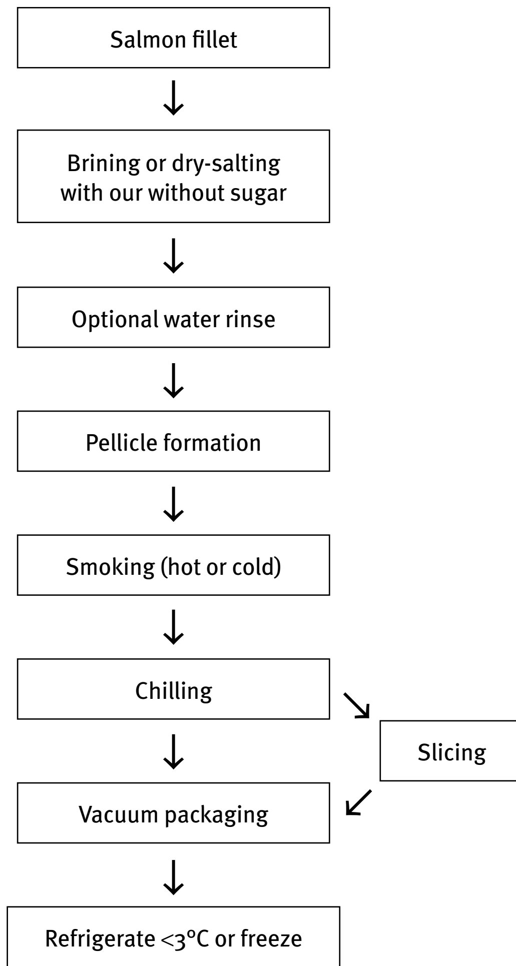

Figure 1. Flow chart for preparing smoked salmon.

The potential for *S. aureus* contamination in smoked fish processing can only occur with direct human handling. Other warm-blooded animals as sources of *S. aureus* in commercial processing plants are nonexistent. Seafood processing steps where contamination may occur are during hand filleting, trimming, brining, pellicle development (Eklund et al. 2004, Himelbloom et al. 2008), filling racks, product slicing, and packaging (Fig. 1).

Twelve reports outside the United States show aquatic foods that contained various levels of *S. aureus* (Table 1). In some cases, these contamination levels would meet or exceed the Food and Drug Administration guideline limit of 10^4 *S. aureus* cells per g for safe seafoods (USFDA 2001). Recommended limits for *S. aureus* in seafood can be subdivided into good quality versus minimum acceptability under international standards (Table 2). The continued presence of *S. aureus* in various seafoods is shown in several studies (Adesiyun 1984b; Sanjeev et al. 1985, 1986; Sokari 1991; Wieneke et al. 1993; Oh et al. 2007; Simon and Sanjeev 2007) although the focus of these reports was on enterotoxin typing and sources rather than bacterial contamination levels.

Staphylococci can be found in retail smoked freshwater fish products in Spain but the incidence and levels of *S. aureus* were reported as minor (González-Rodríguez et al. 2002). High levels of *S. aureus* (10^4-10^5 colony forming units per g) have been determined in Alaska Native smoked salmon strips (Himelbloom et al. 1996, Eklund et al. 2004) and a simulated process (Himelbloom and Crapo 1998, Paranjpye et al. 2004); however, no analysis was conducted to determine enterotoxin type and quantification in these products.

Factors for *S. aureus* growth and enterotoxin production

Bacteria have minimum, optimum, and maximum temperature ranges for growth. Nutrients are necessary for growth and single cell division. Other environmental factors are pH and atmosphere conditions regarding the presence or absence of oxygen. If the growth factors are satisfactory, time is the final factor that determines the rate of a bacteria population to develop. Within or on food, water activity values, presence of bacterial inhibitors, and types of competitors for nutrients will affect *S. aureus* population levels. The temperature range for growth of *S. aureus* is 7-48°C (45-118°F) with an optimum of 37°C (99°F) (Baird-Parker 2000). Enterotoxin production occurs over the temperature range of 10-46°C (50-115°F) with an optimum of 40-45°C (104-113°F) (Baird-Parker 2000). The a_w range for *S. aureus* growth is 0.83 to >0.99 with an optimum of >0.99, while foods at the lower a_w values provide a competitive edge for *S. aureus* against other bacteria (Smith et al. 1983). Enterotoxin is produced under the a_w range of 0.86 to >0.99 and the upper value is optimum (Smith et al. 1983). In an experimental seafood system, shrimp slurries, enterotoxin is not produced until the a_w is at least 0.93-0.95 (Troller and Stinson 1975). A combination of reduced a_w and low storage temperature forestalls *S. aureus* growth and thermonuclease production in smoked snoek (Theron and Prior 1980). Staphylococci are salt-tolerant bacteria and *S. aureus* cells grow in the absence (optimum) or up to 20% salt (Smith et al. 1983) which includes salted or salt-cured fish (Huss and Valdimarsson 1990, Sanjeev and Surendan 1996). Enterotoxin production occurs in bacteriological media containing up to 10% salt but is produced optimally in its absence (Smith et al. 1983).

Controlling *S. aureus* in smoked seafoods

A couple of simple procedures can alleviate concerns caused by potential risks from *S. aureus*. Contamination of ready-to-eat products can be prevented through the use of latex gloves (Himelbloom et al. 1996, Himelbloom and Crapo 1998, Autio et al. 2004, Simon and Sanjeev 2007) or utensils to reduce excessive human hand contact (ICMSF 2000). Open-air markets have been implicated in direct transfer of *S. aureus* during handling between traders and customers of ready-to-eat cooked, smoked, dried, or fried fish and shellfish (Abeyta 1983, Adesiyun 1984a, Saddik et al. 1985, Sokari 1991). Quality assurance managers need to remind person-

Table 1. Aquatic foods containing various levels of *Staphylococcus aureus*.

Fish product (country)	Lab analysis	Risk	Reference
Smoked and dried fish (Nigeria)	*S. aureus* >10^6/g; 37% of strains toxin-positive; enterotoxins A to E present	Post-process (smoked on open fire to dryness) contamination at markets and enterotoxin confirmation before consumption	Adesiyun 1984a
Smoked shad and carp (Iran)	22% of samples >10^4/g	Potential enterotoxin formation before consumption	Basti et al. 2006
Smoked and charred herring (Finland)	15% of summer retail samples >10^3/g	Potential enterotoxin formation before consumption	Hirn and Hirvelä 1983
Smoked and charred herring (Finland)	3% of samples = 10^2-10^6/g	Potential enterotoxin formation before consumption	Korkeala and Pakkala 1988
Smoked fish (Canada)	93% of retail samples averaged 10^3 -10^4/g	Potential enterotoxin formation before consumption	Dillon et al. 1992
Smoked and dried herring (Ghana)	*S. aureus* = 10^3 -10^4/g	Potential enterotoxin formation before consumption	Lu et al. 1991
Fresh and smoked fish (India)	*S. aureus* = 10^2-10^3/g	Post-process contamination at markets	Vishwanath et al. 1998
Raw and cooked fish and shellfish (Egypt)	47% of raw fish, 76% of raw shrimp and 73% of cooked seafood >10^3/g	Post-process (fried or broiled) contamination	Saddik et al. 1985
Pre-cooked and peeled shrimp (Malaysia)	34% of samples >10^3/g	Post-process contamination	Beckers et al. 1981
Raw fish and crustacean meat (Brazil)	20% of samples = 10^1 to >10^4/g; 67% of shellfish meats >10^4/g	Potential enterotoxin formation before cooking	Ayulo et al. 1994
Frozen prawns and fish (India)	20-33% of samples = 10^2-10^3/g	Post-process contamination	Simon and Sanjeev 2007
Raw fish fillets (Netherlands)	4% of samples >10^3/g	Potential enterotoxin formation before cooking	van den Broek et al. 1984

Table 2. Recommended microbiological limits for *Staphylococcus aureus* in seafoods.

Product	Number of samples (n)	Good quality, minimum limits (*m*)	Marginally acceptable, maximum limits (*M*)	Maximum number between limits ($m < c < M$)
Fresh and frozen fish	5	10^3/g	10^4/g	2
Cold-smoked fish	5	10^3/g	10^4/g	2
Pre-cooked breaded fish	5	10^3/g	10^4/g	1
Frozen raw crustaceans	5	10^3/g	10^4/g	2
Frozen cooked crustaceans	5	10^3/g	–	0
Cooked, chilled and frozen crabmeat	5	10^3/g	–	0

Adapted from ICMSF (1986).

nel to avoid contacting skin, nasopharynx emissions, and hair while gloved since the effectiveness of this physical barrier will be compromised and potentially transfer the bacterial pathogen to seafood products. Pasteurization can inactivate only the bacterium but not the heat-stable enterotoxin. Mild heat treatment, 60°C (140°F) for 1-6 min, can inactivate the cells before enough are present for enterotoxin to be detected (Baird-Parker 2000, ICMSF 2000). This range of *D*-values, or the time to reduce the viable cell population by 90%, can be reduced ten-fold by increasing the pasteurization temperature by one z-value equaling 6-10°C (43-50°F) (Baird-Parker 2000, ICMSF 2000). Two other approaches are to process the seafood to reduce the a_w below 0.85 or store the ready-to-eat products below 7°C (45°F).

Conclusions

The food pathogen, *S. aureus*, is an easily controllable pathogen for the smoked seafood industry. Food-borne illnesses from consumption of *S. aureus*-contaminated smoked seafoods are considered unlikely situations due to the implementation of good manufacturing practices and hazard analysis and critical control point plans.

Acknowledgments

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A Review: Processing Parameters Needed to Control Pathogens in Cold Smoked Fish

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Introduction

In 1998, the Food and Drug Administration (FDA) signed a five year contract with the Institute of Food Technologists (IFT) to provide scientific review and analysis of issues in food safety, food processing, and human health. The 2001 report is IFT's response to Task Order 2: Processing Parameters Needed to Control Hazards in Cold Smoked Fish (IFT/FDA 2001a).

Members of the expert panel included
Dr. Frank Busta, Panel Chair and Senior Science Advisor to IFT
Dr. Gleyn Bledsoe, Northwest Indian College
Dr. George Flick, Virginia Tech
Dr. Lone Gram, Danish Institute for Fisheries Research
Mr. Daniel Herman, National Fisheries Institute (NFI)
Dr. Michael Jahncke, Virginia Tech
Dr. Donn Ward, North Carolina State University

This paper provides a summary of the 2001 IFT/FDA report, and gives examples (not all inclusive) of research conducted since the report that addresses many of the research needs identified in the report.

Charge from FDA to IFT

The IFT shall review the scientific literature, shall consult with academic experts, and shall consider the requirements of other government bodies to address the following specific questions (IFT/FDA 2001a):

- Are times and temperatures for cold smoking conducive to outgrowth of pathogens and histamine producing species of bacteria? What is the range of time and temperatures used by the industry during cold smoking? Which pathogens are of concern?
- Review processing parameters that may contribute to *L. monocytogenes* contamination of the product, e.g., incoming product, chlorination of rinse water, injection brining systems, recirculation of brine, etc. The contractor shall provide information on the various pathogens that might be expected to be present on seafood products. These pathogens would include, but are not limited to: *L. monocytogenes*, *S. aureus*, *C. botulinum*, organisms capable of producing histamine in scombroid species and any other organism that may serve as a foodborne pathogen. The contractor shall do an in-depth review of how these organisms are inhibited (or not inhibited) in smoked fish products and define the critical control points of importance to each of them.
- The contractor shall do an in-depth review on the processing parameters that may contribute to *L. monocytogenes* contamination of product, e.g., incoming product, chlorination of rinse water, injection brining systems, recirculation of brine, etc.
- The contractor shall do an in-depth review on the options available to eliminate or inhibit those organisms of public health concern in smoked fish products. The contractor shall evaluate the various preservatives (e.g., the inhibitory effect of wood smoke, liquid smoke, salt, nitrite, and sodium lactate) used alone or in combination and the levels needed as appropriate inhibitors to pathogen growth. The contractor shall include the influence of the time and method of application of such preservatives. In addition, the contractor shall evaluate the suitability of other controls (e.g., pH, water activity, competitive microflora) on the prevention of outgrowth during processing and during subsequent finished product storage)
- The contractor shall provide information on recommended levels of heat or preservatives, alone or in combination that processors can use to establish critical limits for processing a cold smoked product that is free from *L. monocytogenes* and bacterial toxins. The contractor shall review appropriate corrective actions that can be taken when critical limits are exceeded.
- The contractor shall provide information on how processors can validate the adequacy of the above levels in their processing systems and how to verify that their process is adequate on an ongoing basis.
- The contractor shall evaluate the various packaging options (e.g., oxygen permeable packaging, vacuum packaging, modified atmosphere packaging, controlled atmosphere) and their effect on the inhibition of spoilage bacteria and the outgrowth of pathogens. The contractor shall define the term oxygen permeable packaging as it relates to inhibiting the outgrowth of *C. botulinum* and other pathogens (e.g., what characteristics must be present for a product to be considered

"air" packaged). Products packed in high barrier film without a vacuum being pulled may become anaerobic rapidly due to the growth of aerobes in the product and the subsequent production of carbon dioxide.

- The contractor shall evaluate methods to control *L. monocytogenes* on the incoming product. Are there good vessel/harvest handling practices or microbiological monitoring procedures that will prevent contamination of incoming product with *L. monocytogenes*. What are these practices and/or procedures.
- While the scope of this task order is specific to cold smoked fishery products, the contractor shall provide any information in the literature on hot smoked fish that is germane. If the contractor finds sufficient information demonstrating that the time and temperature of the hot smoke is inadequate to eliminate *L. monocytogenes* from hot smoked products, it shall be noted in the review (IFT/FDA 2001a).

The approach to this topic by the expert panel was to review the literature specifically addressing pathogens that may contribute to pathogen growth and reviewed options available to eliminate or inhibit foodborne pathogens in smoked fish products. An in-depth review of safety concerns of current fish harvesting and handling procedures as well as an evaluation of packaging options and their influence on survival or growth of the organisms of concern was also conducted. The panel focused on the most significant and likely to occur hazards in cold smoked products specifically *Listeria monocytogenes*, *Clostridium botulinum*, human parasites, and biogenic amines. In addition to reviewing the scientific literature, the panel contacted other academic and regulatory experts and smoked fish industry members for their input on harvesting, handling, and processing procedures, as well as information on packaging options currently used. A survey was also sent out by the National Fisheries Institute (NFI) to their members who process smoked fish products.

IFT/FDA report conclusions (IFT/FDA 2001a)

Listeria monocytogenes

Given the ubiquitous nature of *L. monocytogenes*, the lack of listericidal steps in the cold smoking procedure, and the ability of the organism to become established in the processing environment and re-contaminate products, it is not possible to produce cold smoked fish consistently free of *L. monocytogenes*. This is not unique to cold smoked fish because this microorganism can be isolated from a wide range of ready-to-eat (RTE) foods.

By adhering strictly to good manufacturing practices (GMPs) and good hygienic practices (GHPs) it is possible to produce cold smoked fish with low levels of *L. monocytogenes*, preferably at <1 cell per g at the time of production.

1. Growth of *L. monocytogenes* in naturally contaminated fish products is significantly slower than predicted by models (using combinations of pH, NaCl, temperature, and lactate) and inoculation studies.
2. Prevention of growth of *L. monocytogenes* in cold smoked fish cannot be guaranteed not to occur using current combinations of NaCl and low temperature; however, growth can be prevented by freezing, by addition of certain additives (for example, nitrite), or by use of bioprotective bacterial cultures.
3. If the organism cannot be eliminated and growth-inhibiting steps are not introduced, the hazard can be controlled by limiting shelf life (at 4.4°F) to ensure that no more than 100 cells per g are present at time of consumption. Time limits may need to be established by each processor because the time limit should reflect the initial level of the organism in freshly produced product.
4. Some countries, such as Australia, warn pregnant women about listeriosis and offer a list of food items to be avoided during the pregnancy. Labeling cold smoked fish as well as other RTE foods in this risk category, indicating that these products may constitute a health hazard for immunocompromised individuals and pregnant women could be considered.
5. There is no control point during the cold smoking process that will guarantee the elimination of *L. monocytogenes* on the final product; however, the occurrence of *L. monocytogenes* on finished cold smoked fish products of processors can be minimized by (1) obtaining the primary product from known sources (for example, those with a history of non-contaminated fish); (2) following strict adherence to GMPs to prevent recontamination during processing; and (3) inhibiting growth of any survivors by marketing the product frozen, or by using salt and other preservatives that can inhibit growth at refrigerated temperatures.

Clostridium botulinum

1. Psychrotrophic *C. botulinum* occurs naturally in the aquatic environment, so its presence in low numbers on fresh fish must be anticipated. Spores may also be isolated infrequently from cold smoked fish, although numbers, if present, are low. Given this low number, the probability of germination and toxin production is low but present.
2. Experiments with naturally contaminated hot-smoked fish produced from fish with high levels of *C. botulinum* show that toxin may be formed under conditions of temperature abuse.
3. Toxin production by psychrotrophic *C. botulinum* is controlled with a combination of a moderate level of NaCl (3.5% NaCl WPS) and storage at chill temperature (<4.4°C, <40°F) for at least 4 weeks. Based on the scientific data and because commercially produced

cold smoked fish has never been reported as a source of botulism, it is reasonable to conclude that the salt and cold keep the hazard under adequate control.

4. Based on a range of model studies in broth and inoculation studies with hot or cold smoked fish, it can be concluded that a combination of 3.5% NaCl (as water phase salt) and chill storage (4.4°C, 40°F or lower), allowing for short time periods of elevated temperature up to 10°C (50°F), will prevent toxin formation in reduced oxygen packaging of cold smoked fish for several weeks beyond its sensory shelf life.
5. As a general safeguard, salting to 3.5% for chilled stored cold smoked fish is essential for reduced oxygen packaged (ROP) cold smoked fish. In addition, the requirements for chilling with a sufficient salt concentration are an option for consideration in national or international regulations (for example, E.U. directives).
6. For air-packaged products, levels of NaCl can, theoretically, be reduced; however, scientific data that support this argument do not exist and are needed before any reduction is recommended. Even when not packed under vacuum or modified atmosphere, pockets of anaerobic conditions may be created where slices of fish overlap or where aerobic spoilage bacteria consume the oxygen present.
7. To control *C. botulinum* growth and toxin production in ROP products the following considerations are indicated: (1) A minimum 3.5% water phase salt concentration in the thickest part of the fillet for vacuum or modified atmosphere packaged fish, or a combination of at least 3% water phase salt and a nitrite level of 100-200 ppm is necessary for the control of *C. botulinum* growth and toxin formation (Note: nitrite is not allowed in products sold in Europe, and is only allowed in the United States for sablefish, salmon, shad, chub, and tuna); (2) Packages containing refrigerated, cold smoked fish should be labeled, "Keep refrigerated at 40°F (4.4°C) or below; (3) Packages containing frozen, cold smoked fish should be labeled, "This product must remain frozen until thawed at refrigeration temperatures and shall not be refrozen"; and (4) Products should not be packaged in reduced oxygen packaging by the retailer.

Biogenic amines

1. The majority of species that are cold smoked have not been identified by the scientific community as causing scombrotoxin illness. Therefore, the risk of foodborne illness is limited in the majority of cold smoked products available in the marketplace.
2. Usually, only relatively high and sometimes controversial concentrations of histamine have resulted in illness. The contribution of other biogenic amines to the onset of symptoms is not well understood.
3. Most scombrotoxin results from extrinsic, rather than intrinsic, spoilage through the growth of certain bacteria, generally members of the family Enterobactericae. Some bacteria are capable of producing greater quantities of decarboxylase enzymes than others.
4. Certain processing operations, such as freezing, salting, or smoking may be capable of inhibiting or inactivating biogenic amine-producing microorganisms; however, microorganism growth with potential toxin production may occur after thawing and post-processing.
5. Under certain conditions addition of lactic acid-producing microorganisms suppresses the growth of biogenic amine-forming microorganisms.
6. Vacuum packaging does not prevent growth of biogenic amine-forming microorganisms.
7. While biogenic amine-forming microorganisms may grow at refrigeration temperatures, generally the minimal temperature for growth is lower than the minimal temperature for toxin production.
8. The most effective methods of preventing biogenic amine formation are handling and processing under sanitary conditions, rapid cooling of the fish, and continued refrigeration from harvest through consumption.
9. To minimize the level of biogenic amines in species susceptible to histamine formation, temperature control is important throughout the process, particularly during the storage and transportation before cold smoking, the cooling step, and the final product storage, distribution, retail, and consumer steps. The temperatures required for the control of *C. botulinum* may be appropriate to control production of biogenic amines.
10. Much of the published scientific research on scombrotoxin utilized fish samples obtained from processing facilities and retail food stores. Only a limited number of studies followed samples from harvest through analysis. Also, sensory analyses were not always incorporated into microbiological and analytical chemical studies. There is a lack of reports describing comprehensive and integrated projects.

Parasites

1. Some of the fish species used for cold smoked processing are either intermediate or final hosts to parasites. For this reason, assuring the harvesting of parasite-free fish in the wild is difficult.
2. Some aquacultured fish are considered free of parasites (if their feeding regime has not been supplemented with raw fish) because their diet can be controlled using net-pens, closed recycled systems or an equivalent system, and commercially pelleted diets; consequently, these control measures must be

carefully considered and applied. An analysis of the potential control points for parasites in aquacultured fish is beyond the scope of this report.

3. Freezing raw fish prior to smoking remains the most effective way to insure that viable parasites are not present in cold smoked products consumed by the public. It is essential, therefore, that raw fish potentially containing viable parasites be frozen and held in that state for a period of time that assures destruction of all viable parasites in that fish species.

IFT/FDA report research needs (IFT/FDA 2001a)

The following is a list of research needs identified by the panel.

Listeria monocytogenes

1. Conduct epidemiological investigations to determine if and to what extent cold smoked fish is involved in cases of listeriosis. Despite prediction of a risk, only a limited number of cases have been associated with cold smoked fish.
2. Assess virulence potential of *L. monocytogenes* isolated from cold smoked fish.
3. Measure behavior of *L. monocytogenes* in naturally contaminated products. *Listeria monocytogenes* appears to grow more slowly and to lower numbers than anticipated based on model predictions and inoculation trials. An understanding of which factors cause these differences may be used to design appropriate control measures in the product.
4. Determine the robustness and applicability of alternative growth inhibitory measures such as bioprotective cultures, bacteriocins, lactate, and others.
5. Determine how *L. monocytogenes* becomes established in smoke houses and processing facilities. Several studies show that particular DNA types become established in niches in the processing environments. Research is needed to evaluate what parameters determine which types reside—whether it is particular adhesion properties, or particular resistance properties, or other factors.
6. Investigate the source of contamination in smoke houses and processing environments in order to introduce procedures specifically targeted at eliminating or limiting introduction of the organism.
7. Identify GMP practices that would minimize the contamination and growth of *L. monocytogenes.*
8. Determine the effectiveness of intervention strategies to reduce or eliminate *L. monocytogenes*, such as using chlorinated water to thaw and rinse incoming fish, and for rinsing fish following the brining operation.
9. Develop cleaning and disinfection procedures targeted at adhered or established cells for removal of *L. monocytogenes* from surfaces.
10. Determine if particular types of surfaces reduce numbers of adhering *L. monocytogenes* or if particular treatments (that is, spraying with lactic acid bacteria or lactate) can reduce surface contamination by minimizing adhesion and biofilm formation.
11. Evaluate the robustness and sensory acceptability of the various procedures under investigation (that is, bioprotection, lactate, and so on) for the elimination of the possibility of growth in the product.
12. Determine the effect of post-processing methods such as irradiation and high pressure to eliminate *L. monocytogenes* in cold smoked fish.

Clostridium botulinum

1. Evaluate growth and toxin production in naturally contaminated cold smoked fish products to validate models and predictions for growth and toxin production.
2. Determine the influence of redox potential, various concentrations of trimethylamine oxide (TMAO), and NaCl on toxin production by psychrotrophic *C. botulinum* in gadoid and non-gadoid species.
3. Determine the potential facilitation by TMAO on formation of nitrosamines, if nitrite is added, during cold smoking.
4. Identify processing conditions and gas transmission rates of films under various time and temperature conditions for products to be considered "air packaged." Determine the oxygen transmission rate (OTR) needed for a product with 2.5% salt concentration to provide equivalent safety compared with cold smoked reduced oxygen-packaged (ROP) products.
5. Conduct challenge studies on air-packaged, cold smoked fish in films with OTRs between 7,000 and 10,000 cc per m^2 per 24 h and compare to unpackaged cold smoked fish.
6. Establish minimum water phase salt concentrations required to inhibit growth and toxin formation by *C. botulinum* in air-packaged and unpackaged cold smoked fish.
7. Determine the shelf life of the product relative to product quality as well as safety under different packaging methods and storage temperatures.
8. Determine appropriate sell-by dates and evaluate the use of time-temperature indicators to ensure a safe product.

Biogenic amines

1. Determine the influence of ROP of products on the inhibition of biogenic amine production by gram-negative bacteria.
2. Define the minimum temperatures for growth and biogenic amine production of biogenic amine-forming microorganisms.
3. Identify practical temperatures that would minimize the levels of biogenic amines in all steps of the production chain and in the final product.
4. Determine the effect of salt and redox potential on the formation of biogenic amines on the final product.
5. Determine the impact of the inter-relationship(s) among histamine, putrescine, cadaverine, and perhaps other biogenic amine concentrations in scombrotoxin and their effects on subsequent host responses.
6. Investigate the effects of various cold smoked fish processes (water phase salt concentrations, process times and temperatures) on biogenic amine formation.
7. Identify practical methods for cold smoked fish processors to determine the histamine/scombrotoxin risk in the raw material used for smoking.
8. Apply new processes, such as irradiation, modified atmospheres, or high pressure, to reduce specific groups of microorganisms to determine if control of those responsible for biogenic amine formation reduces the hazard.
9. Evaluate the effects of harvesting methods and post-harvest handling practices on biogenic amine formation under varying environmental conditions.
10. Identify specific methods for representative and effective sampling and for accurate and precise analysis of biogenic amines.

Parasites

1. Describe possible alternative freezing procedures that are or could be effective for inactivation of various fish parasites.
2. Establish the kinetics and lethal effect of specific regimes of freezing on various fish parasites.
3. Evaluate alternative processing procedures, such as high pressure and X-ray or e-beam irradiation for control of various fish parasites.
4. Investigate the possible human health risks of allergic reactions due to parasite antigens remaining after freezing the fish to inactivate the live parasites.

Pertinent research 2001-2007

Many of the research needs identified in the IFT/FDA report have been addressed during the six years following the report. The majority of the studies have focused on *L. monocytogenes*, but research has also focused on *C. botulinum* toxin formation in refrigerated reduced oxygen packaged (ROP) fish; understanding and controlling histamine formation in tuna, and to a more limited extent destruction of parasites in fish. Listed below are some of the research activities conducted during the past six to seven years. These are not all inclusive, but they provide a good overview of pertinent research.

Listeria monocytogenes

Surveys have been conducted on the prevalence of *Listeria monocytogenes* on seafood products including seafood salads and smoked fish products (Gombas et al. 2003). In addition, the FDA and Food Safety and Inspection Service, U.S. Department of Agriculture (FSIS/USDA) published a risk assessment for *L. monocytogenes* in ready-to-eat (RTE) foods (FDA/USDA 2001). Research has also been conducted on mathematical modeling of *L. monocytogenes* in RTE meat products (Dalgaard et al. 2004, Carrasco et al. 2007, Mejlholm and Dalgaard 2007).

Extensive research has been conducted on how *L. monocytogenes* becomes established in smoke houses, and likely niche areas in smoke houses and processing environments where *L. monocytogenes* is commonly found (Norton et al. 2001, Thimothe et al. 2004, Hansen et al. 2006). In addition, research on identification of persistent strains of *L. monocytogenes* found in smoked fish processing plants has been conducted (Wulff et al. 2006). Potential treatment options such washing incoming raw material with potable water containing chlorine (<10 ppm), washing raw fish with calcium hydroxide to reduce or eliminate *L. monocytogenes* (Jahncke et al. 2004), and freezing finished product or adding lactates to finished product to control growth of *L. monocytogenes* have been investigated (Jahncke et al. 2004, Vogel et al. 2006). Research was conducted on the effectiveness of high pressure processing to control *L. monocytogenes* on incoming raw fish and finished product. The results indicate that high pressure processing was unable to prevent growth of all strains of *L. monocytogenes*, and the process had a marked negative effect on the color and texture of raw fish and on finished cold smoked salmon product (Lakshmanan and Dalgaard 2004).

Guidance has been provided to industry on how to apply good manufacturing practices (GMPs) and good sanitation practices to control contamination of the processing environment, food contact surfaces, and finished product by *L. monocytogenes* (Gall et al. 2004). Recommended guidelines were provided to industry on how to sample and test the processing environment, and how to sample and test raw product and finished product samples for *L. monocytogenes* (Scott et al. 2005). Suggestions on how to train employees in good cleaning and good sanitation practices, and how to implement and follow GMPs have also been provided to the industry (Hicks et al. 2004).

Clostridium botulinum

The National Advisory Committee on Microbiological Criteria for Foods (NACMCF) published an article on scientific parameters needed to establish safety-based use-by dates for RTE foods (NACMCF 2005). Vestergaard (2001) published research addressing how to set up microbial challenge studies for *C. botulinum*, and Cates et al. (2007) evaluated consumer storage practices of RTE foods. Although the research was not on smoked fish, recent studies on film oxygen transmission rates (OTRs) and production of botulinum toxin in inoculated ROP refrigerated raw fishery products has also been conducted (Arritt et al. 2007, Rheinhart 2007). Their research demonstrated that spoilage and toxin formation in refrigerated raw fish (i.e., flounder [*Paralichthys dentatus*] and croaker [*Micropogonias undulatus*]) packed under ROP conditions using films with OTRs less than 10,000 cc per m^2 per 24 h at 70°F were comparable with refrigerated raw fish packed under films with an OTR of 10,000 cc per m^2 per 24 h at 70°F. In addition, Dr. Michael Doyle, Univ. of Georgia, is currently conducting research on inoculated pack studies with *C. botulinum* on raw salmon packaged under films with different OTRs (M. Losikoff, Food and Drug Administration, Center for Food Safety and Applied Nutrition, College Park, Maryland, pers. comm., 2007). Information from studies such as these can be used by the FDA as they re-evaluate their regulations on safety issues for refrigerated ROP fishery products including smoked fish.

Biogenic amines

Research on histamine formation in cold smoked scombrotoxic fish was conducted in Denmark (Emborg and Dalgaard 2006). Major research efforts in the United States have focused on the Pacific longline tuna fishing industry. U.S. research efforts focused on the effects of harvest methods, vessel handling, and storage practices, and onboard gilling/gutting to control histamine formation in large fresh tuna (>20 lbs.) (Kaneko 2000, 2004; NSIP 2005).

Parasites

Although the hot fish smoking process is adequate to destroy parasites, the cold smoking process does not destroy parasites (IFT/FDA 2001a,b). However, the FDA Fish and Fishery Products Hazards and Controls Guidance Document provides information to processors on how to kill parasites in fish by freezing (FDA 2001). A new edition of the guidance document is scheduled for publication sometime in 2008.

In 2004, Dong et al. evaluated if high pressure processing could be used to inactivate *Anisakis simplex* in fish destined for the sashimi market. Her results demonstrate that the pressures needed to kill 100% of the parasites also resulted in significant whitening in the color of the salmon.

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Bacteriophage Control of Pathogens in Foods

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Introduction to bacteriophages

Bacteriophages are viruses that attack bacteria. They attach to bacteria, and inject their DNA or RNA. Then they use the bacterial cell components to make new virus particles. They are host-specific organisms, and only affect a certain type of bacterium; they will not infect plant or animal cells, which is an advantage to us.

There are two types of bacteriophages: virulent or lytic types, and temperate or lysogenic types. This becomes important when talking about controlling pathogenic bacteria in food. Lytic phages infect a bacterial cell and use the cell components to make more bacteriophages. They produce enzymes called endolysins that lyse the cell from within, and this releases the phages.

The temperate or lysogenic phages are a little different. They enter the cell and integrate into the chromosome of the cell. They stay dormant until some stress activates them. Then they come out of the chromosome and in doing so they often pick up bits of DNA from the bacterial cell—so lysogenic phage use is not a good method for controlling pathogens in food.

Using lytic activity in food safety

Endolysins are phage-encoded cell-wall lytic enzymes. Endolysins are synthesized late in the stage of viral multiplication because they allow the viruses to escape from the bacteria. When the phages lyse the cell, they release 50 to 200 new phage particles; they can quickly amplify themselves.

An increasing amount of research is being done on phage control of pathogens such as *Salmonella* (Leverentz et al. 2001), *Listeria monocytogenes* (Gaeng et al. 2000, Loessner 2005, Carlton et al. 2005, Turner et al. 2007, Vermeiren et al. 2007), *E. coli* O157:H7 (FSIS 2006, Sheng et al. 2006), *Campylobacter* (Wagenaar et al. 2005), and *Enterobacter sakazakii* (Kim et al. 2006). Phages that attack these pathogens produce endolysins, and researchers are interested in what these endolysins do to bacterial cells.

Studies have shown the effectiveness of a lytic phage, A511, against *Listeria monocytogenes* (*Lm*). Endolysins were mixed with *Lm* and within two minutes almost all the *Listeria* were gone, and 30 seconds later there were no *Listeria* cells left (Loessner 2005). Thus, this can be a rapid process.

Particular phages have specific reactions. Phage A511 reacts with *Listeria monocytogenes* first through lysis by the phage, and then by lysis by the endolysin. To enumerate a phage, a lawn of bacteria is made into a plate of agar; then the bacteriophage-containing agar is overlaid. Where lysing occurs there are clear spots. These are counted as plaque-forming units (pfu). A halo effect indicates further lysing around the area, showing that when the endolysin is released upon lysis of the *Listeria* cells, it diffuses out and lyses additional cells.

Recombinant *E. coli* cells were produced and treated with chloroform to make them leaky and allow the endolysin proteins to leak out of the cells (Loessner 2005). Then they were overlaid with a *Listeria*-containing soft agar. This caused the endolysin to leak out of the *E. coli*. It did not cause lysing of the *E. coli* but rather caused the lysing of the *Listeria* in the overlay. An endolysin, which is meant to lyse a cell from within, can also lyse a cell from without when the cell wall is accessible. This can be useful in controlling pathogens in food.

Bacteriophage control of food-borne pathogens

Cheese

One possible use of lytic activity is in production of starter cultures for dairy products that have the activity to lyse the cells of pathogens of concern.

Cells of *Lactobacillus lactis*, that were not transformed in any way, were compared to recombinant strains that were developed with an endolysin that is secreted by the *L. lactis* cells, which can lyse *Listeria monocytogenes* (Gaeng et al. 2000, Turner et al. 2007). When they are overlaid with a layer of *Lm* we get lysis of *Lm*. We can make cheese with starter cultures that are active against *Lm*; this has great potential.

Cooked chicken

Other studies show the effect of phage P100 on *Lm* on cheese (Carlton et al. 2005) and cooked chicken (Table 1). *Lm* grows over 21 days at refrigeration temperatures. When the phage

Table 1. **Growth of a three-strain *L. monocytogenes* cocktail (log_{10} cfu per g) on vacuum packaged cooked chicken fillet at 7°C without P100 (LIS) and with P100 (LIS+P100) at a dose of 1×10^7 pfu per cm^2 (Vermeiren et al. 2007).**

Time (days)	LIS	LIS + P100
0	1.00	1.00
7	2.46	2.04
14	4.62	1.85
21	4.32	1.00

Table 2. Phage reduces *Campylobacter jejuni* colonization in broilers (Wagenaar et al. 2005).

	Preventive measure	Therapeutic measure
Time broilers infected with *C. jejuni*	Day 4 of a 10-day phage treatment	
Time broilers treated with phage		5 days after *C. jejuni* colonization
Initial observation	2 log reduction in *C. jejuni* and delayed colonization in treated group	3 log decline of *C. jejuni* in treated group
Final observation at end of experiment	Comparable *C. jejuni* in treated and control group	1 log lower in treated than control group

plus *Lm* is introduced there is a small amount of growth, but ultimately the phage beats the *Listeria* down.

Fruit

Further studies have looked at the effect of bacteriophages against *Salmonella enteritidis* (SE) on fresh cut fruits (Leverentz et al. 2001). Four phages were combined to attack SE on honeydew melon slices. The results were temperature dependent, reducing *Salmonella* by 3.5 logs at 5 or 10°C but only 2.5 logs at 20°C. But the same phage treatment did not work on apple slices, showing that phages are sensitive to low pH. In the apple study, not only was the *Salmonella* not lysed but the phage disappeared; the acid may have inactivated the phages. For low pH fruits some new phages must be isolated.

Chicken broilers

Control of colonization of chicken broilers by *Campylobacter jejuni* was studied using two approaches—one preventive and one therapeutic (Table 2). Both approaches had limited success, reducing *C. jejuni* by 2 or 3 logs but not completely eliminating it (Wagenaar et al. 2005). This indicates that phage treatment is a promising alternative for reducing *C. jejuni* in broilers, but additional work is needed to find the conditions that will improve the effectiveness of phage therapy, for example, large-scale propagation of phages.

Infant formula

Phages were effective in inhibiting growth of *Enterobacter sakazakii* in reconstituted infant formula. This organism has caused severe problems for premature and very ill babies who were fed contaminated formula that had been held for extended periods. Comparing two different phages, ESP1-3 and ESP732-1, held at two different temperatures, 12°C and 24°C (Kim et al. 2006), better results in inhibiting growth of *E. sakazakii* were obtained at the higher temperature. Both phages were less effective at 37°C, but phage ESP 732-1 at 9 log resulted in complete inhibition.

Other phage uses

The use of bacteriophage is not new. Back in the 1930s and 1940s phages were used extensively to treat bacterial infections in humans (Sulakvelidze and Kutter 2005). However, although some were highly effective, the therapeutic effectiveness was inconsistent because there was not good information on the correct amounts or delivery to the sites to attack bacteria. Bacteriophages were not used much after the development of antibiotics. Using phage required isolating the bacterium, knowing the specific target organism, then isolating a phage that would attack that particular bacterium. The broad-spectrum antibiotics were easier to apply. The recent return to the use of phage is a reaction to the increase in antibiotic-resistant bacteria. Phage therapy is still widely used in Russia and Eastern Europe (Sulakvelidze and Kutter 2005).

Phages can potentially be used

- As an alternative to antibiotics for treating animal diseases in animal production.
- To prevent colonization of animal feed by food-borne pathogens, by adding phages to feed and water.
- To eliminate food-borne pathogens from the exterior of animals, for example as a hide-wash to reduce *E. coli* O157:H7 on cattle.
- To eliminate food-borne pathogens such as *Listeria monocytogenes* in the environment of facilities and on equipment, as in drain colonization.

Limitations to using phages

There are limitations to using phages. Some phages can transfer bacterial genes and transform infected bacteria; phages as food additives require regulatory approval; practical routes of administering the use of phages are needed; and viral stability may influence the effectiveness of phages.

Factors influencing effectiveness of phages

The physiological state of the host and phage concentration influence the effectiveness of phages. Temperatures allowing growth of the pathogen can overcome the effect of the phage. Phages are inactivated at acidic pH—gastric acidity must be neutralized in human phage therapy for oral treatments.

To address the limitation of phage host-specificity, a "cocktail" of several phages can be used. Multiple phages can increase the host range, and also reduce the likelihood that development of resistance will eliminate the effectiveness of the phage preparation. If cocktails are used, resistant bacterial mutants may be lysed by other phages in the cocktail.

Many studies have shown that bacteriophage-resistant mutants can occur with low frequency, and they often

revert to phage sensitivity. New phages active against newly emerged resistant bacteria can be rapidly isolated, whereas identification of a new antibiotic or sanitizer usually takes much longer.

Phage sources

Three companies are currently making bacteriophage. EBI Food Safety, located in the Netherlands (http://www.ebifood-safety.com), makes Listex™ P100 strain. It is a single broad host–range phage, active against *Listeria monocytogenes*, and is generally regarded as safe (GRAS) for *Lm* control at levels up to 10^9 pfu phage per gram of cheese in brie, cheddar, Swiss, and other cheeses that are aged and ripened. EBI sent a letter to the FDA that self-affirmed their GRAS status, and the FDA posted a letter of no objections on their Web site in October 2006 (FDA 2006b).

Intralytix, located in Boston (http://www.intralytix.com), has developed a phage preparation called LMP-102™, which is a cocktail of six phages that are active against *Lm*. They received FDA approval in August 2006 for ready-to-eat meat products (FDA 2006a).

LMP-102 reduces *Listeria monocytogenes* on turkey hot dogs, packaged sliced turkey, and all-beef hot dogs. In all cases the increase of phage reduces the *Lm* proportionally. None of these cases completely eliminated *Lm*, but in each study the *Lm* was reduced 2-3 logs.

Omnilytics, located in Utah (http://www.phage.com/home4.html), has been using phages to treat cow hides for *E. coli* O157:H7. Omnilytics has a history of spraying produce with phages to reduce pathogens, which is very timely considering the recent spinach scares.

Bacteriophage use on seafood

Omnilytics has successfully treated *Salmonella*-infected water in commercial shrimp habitats, especially important in certain multifaceted aquaculture facilities such as those with chicken houses above aquaculture ponds.

EBI Food Safety has indicated that fish processors are a target industry. Because of their European location, cold smoked products are likely to be among their first projects.

Intralytix has expressed a desire to expand application to seafood, particularly cold smoked products, and is looking for an industry partner who would provide products and allow thorough studies to be conducted.

Advantages of bacteriophage use

There are many advantages to using phages. Phages are natural controls for bacteria, and they have no effect on smell, taste, texture, or color of food. In the presence of the target organism, phages increase in number (unlike antimicrobial agents). Phages are host-specific—they affect only target bacteria.

Bacteriophages are ubiquitous in the environment, and are environmentally safe. The total number of phages on earth is estimated at 10^{30}-10^{32}, with more than 100 million phage types (Sulakvelidze and Barrow 2005). Unpolluted water contains ~2.5×10^8 phages per ml, and there are 10 billion in 100 g of soil. On average, there are more than 10 phages per microbe. Poultry products, fruits and vegetables, and cheese sold at retail often contain more than 10^8 pfu per g.

Phages are common in the human mouth, where they are harbored in dental plaque and saliva. In the United States, approximately 3×10^9 coliphages are shed per person per day.

Bacteriophage safety

Phages are considered safe for several reasons. They are host-specific, so they will not infect animals (including humans) or plants. There is no evidence of negative effects in over 80 years of studies on phage-animal and phage-human interaction. Also, phages do not affect normal spoilage microflora that can act as competitive inhibitors for pathogens.

To ensure safety, it is important to use lytic phages only—lysogenic phages can transfer bacterial genes, including virulence factors and antibiotic resistance. When phages are produced using host cells (e.g., *Listeria*), purification steps are critical to remove (1) viable *Listeria* used to culture phages; (2) bacterial components, e.g., virulence factors and lipopolysaccharides; and (3) medium components that could be potential allergens (FDA 2006a).

Will consumers accept phage treatment?

When the FDA announced the approval of LMP100 for meat products, there was a great deal of negative traffic on the Internet. Food & Water Watch has requested a "stay of action and a formal evidentiary public hearing," indicating that the FDA failed to follow guidelines for assessing safety of additives and that the safety assessment studies provided by Intralytics were inadequate. They also indicated that the efficacy studies were inconsistent with the goal of meeting zero tolerance policy for *L. monocytogenes*. Finally, they indicated that the research submitted was not published in a peer-reviewed journal.

Conclusions

The use of phages for control of pathogens in foods and food animals has promise. Phage control of *Listeria monocytogenes* may be particularly useful in ready-to-eat foods that lack a kill step and that support growth, such as cold smoked salmon. Although the "natural" aspects of bacteriophage should appeal to consumers, the addition of live viruses to foods has generated consumer concern.

The industry's next step will be to undertake risk communications and make it clear that these organisms are natural and host-specific and should not be of concern.

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Questions and answers

Question: Why can't phage mutate to infect animal cells?

Answer: A bacteriophage is designed to infect a bacterium relying on the host cellular mechanism. There's very little DNA in a bacteriophage so it would take some serious mutation to produce a mutant that could infect animal cells (it is not likely).

Question: Does your body look at phage as an invading virus?

Answer: You would be consuming a protein that would not be normal to the body so theoretically somebody could develop an allergic reaction, but that is probably not common.

Question: How would you label phages?

Answer: The label would read "bacteriophage preparation." So the consumer would have to be aware and knowledgeable.

Question: What is the use level?

Answer: In cheeses the use level is 10^9 pfu per gram. You need very high levels.

Question: Wouldn't the high levels cause spoilage?

Answer: No; the phages only grow in the bacteria.

Pellicle Formation and Inactivation of *Listeria* and *Staphylococcus* Species in Hot-Smoking of Salmon

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"The newly ingendred juyces, in their own pellicles. Simpson, W. 1669. Hydrol. Chymica 276" (Oxford University Press 2007).

Abstract

Bacteria may resist the inhibitory action of wood smoking by protection under a protein film that develops after brining of fish. When salmon are dried prior to smoking, a pellicle forms and may prevent phenolic and acid compounds in smoke from interacting with bacteria to cause cell death. Our objectives were to characterize the pellicle and to determine survival rates for *Listeria innocua* ATCC 33090 and *Staphylococcus epidermidis* ATCC 14990 (surrogates for *L. monocytogenes* and *S. aureus*) during drying and hot-smoking processes of inoculated salmon. Uninoculated pellicles had higher percentages of protein (46%), fat (4%), ash (5%), and water-phase salt (4%) and lower water activity (0.92) than the underlying tissues. Commercially produced pellicles were analyzed and varied in these components. Pellicle thicknesses averaged 1.3 mm. During a five-stage, six-hour smokehouse program, *L. innocua* and *S. epidermidis* counts declined 4-5 log cycles in the strips in which smoke was added initially. Liquid-smoked strips resulted in a 3 log reduction early in the smokehouse program followed by an additional 2 log reduction. When no smoke was present on the strips, only a 2-3 log reduction occurred. Liquid smoke added at a middle stage showed a 4 log reduction while controls showed 3 log reductions. Wood chip smoke or liquid smoke were necessary to provide additional inactivation of *L. innocua* and *S. epidermidis* in hot-processed salmon strips.

Introduction

A pellicle layer is formed after raw fish flesh has been salted and dried for a period of time during which exposed surface proteins are denatured (Autio et al. 2004). After salmon flesh is brined, the surface is damp, has a slippery feeling, and becomes tacky from brine-eluted proteins (Dillon et al. 1994, Horner 1997, Eklund et al. 2004). The artificial, glossy skin or pellicle forms on the cut surface of fish and this finish helps seal in natural juices and flavors (Dillon et al. 1994, Horner 1997).

Brined fish are placed on racks and dried to enhance pellicle formation, to reduce drying time during smoking, and provide maximum smoke exposure to fish flesh (Dillon et al. 1994). Drying occurs in the smokehouse or kiln without applying smoke and is considered the first stage of smoking (Horner 1997, Autio et al. 2004). During initial steps of smoking, a high rate of drying and heavy deposition of smoke may occur (Dillon et al. 1994). These drying concepts, regarding when to begin smoke deposition, contrast with the recommendation by Eklund et al. (2004) to add smoke to brined fish before a pellicle is formed.

Most smoke components are absorbed optimally by the exposed, wet fish tissue and interstitial water in fish muscle (Horner 1997). The impervious, protective pellicle can form before inhibitory concentrations of smoke constituents are deposited (Poysky et al. 1997, Eklund et al. 2004, Jinneman et al. 2007). The bacteria are theorized to be either embedded in or under the pellicle; when the pellicle is formed before application of generated smoke or liquid smoke, the inactivation of *Listeria monocytogenes* and *Staphylococcus aureus* is markedly reduced in both cold- and hot-smoked salmon (Poysky et al. 1997, Eklund et al. 2004).

Listeria monocytogenes is a problematic pathogen, a ubiquitous contaminant in some raw and processed foods, and a primary concern for processors of smoked fish (IFT 2001, Jinneman et al. 2007). Domestic producers and importers of smoked fish must adhere to the policy of zero tolerance for *L. monocytogenes* (USFDA 2001b); otherwise the products will be recalled from the market. Almost 120,000 kg of ready-to-eat fish and seafood, including approximately 50,000 kg of smoked finfish, were recalled during the past 20 years (Jinneman et al. 2007).

Post-processing contamination by both *L. monocytogenes* and *S. aureus* can occur in hot-smoked fish. The latter pathogen is found in a unique process whereby salmon strips are handled routinely during smoking under ambient temperatures for several days (Himelbloom et al. 1996, Himelbloom and Crapo 1998, Eklund et al. 2004, Paranjpye et al. 2004). Aspects of *S. aureus* in smoked fish were reviewed in these proceedings (Himelbloom 2008).

The purpose of this project was to determine the compositional analysis of pellicles and to monitor the changes of pathogen surrogates inoculated prior to pellicle development followed by hot-processing (no smoke) or hot-smoking of salmon strips. Different smoke applications of inoculated salmon strips were investigated by using a pilot-scale

programmable smokehouse. Pellicles from commercial smokehouses were obtained for comparing the analysis with those conducted experimentally.

Materials and methods

Salmon strips

Freshly harvested Alaska pink salmon (*Oncorhynchus gorbuscha*) were obtained from local processors in Kodiak and brought to the pilot plant in the Fishery Industrial Technology Center (FITC). Fish were headed, gutted, and stored frozen (−30°C). Fillets were prepared from thawed fish and cut into strips, approximately 10 cm × 2 cm × 2 cm, and brined in 5% NaCl (w/v) overnight at 4°C. The ratio of brine to strips was 2:1 (v/w). Strips were removed from the brine and arranged on racks to drain residual liquid for about 1 hour.

Inoculated packs

Overnight cultures of *Listeria innocua* ATCC 33090 and *Staphylococcus epidermidis* ATCC 14990 (both obtained from the American Type Culture Collection, Manassas, Virginia) were grown in Brain Heart Infusion broth (Difco Laboratories, Detroit, Michigan) at 35°C to reach about 10^7 - 10^8 cells per ml. An Eppendorf pipettor (VWR International, Brisbane, California) was used for delivering 100 µl of each culture on drained strips and sterile bent glass rods were used to spread the inocula. The inoculated strips were dried using a portable circulating electric fan to provide air movement of about 5 m per second for 30 min at 25°C.

Smokehouse operation

The racks were placed on a rolling cart and moved into the smokehouse (Model CHU-150, Enviro-Pak, Inc., Clackamas, Oregon) in the FITC pilot plant. The six-hour smokehouse program consisted of five stages: 30 min at 35°C, 60 min at 45°C, 90 min at 55°C, 90 min at 65°C, and 60 min at 70°C. Three experimental studies were conducted: wood smoke added initially vs. control (no smoke); liquid smoke as a 1 min dip in 60% Char-Sol Supreme (Red Arrow, Manitowoc, Wisconsin) solution in deionized water (v/v) before inoculation (Vitt et al. 2001); and a 10% Char-Sol Supreme solution in deionized water (v/v) sprayed on strips (after inoculation) initially or midway in the smokehouse program. At the conclusion of the program, the cart with racks was removed from the smokehouse to equilibrate to room temperature (25°C) for 1 hour. The cart was rolled into the walk-in cooler (5°C) for storage overnight before final products were analyzed. Real-time temperatures at 1-min intervals in the smokehouse were recorded using i-Button dataloggers (Maxim, Dallas, Texas) placed on a smokehouse rack. Temperature data were downloaded to a computer and graphed using Excel 2003 (Microsoft Corp.).

Bacterial sampling and analysis

Pellicle samples were excised aseptically by sterile knife and forceps to remove the external 1-2 mm layer from the strips and collected in sterile sampling bags (Whirl-pak, Nasco, Fort Atkinson, Wisconsin). The inocula, samples from pre-smokehouse (post-drying), three of the intermediate smokehouse stages (#2, 3, and 4) and final products were analyzed for *Staphylococcus* and *Listeria* bacterial counts. A minimum of six strips were sampled to provide a composite of ~0.5 g for the post-dried pellicles and ~5 g for intermediate and final products. Samples were serially diluted using sterile 0.1% (w/v) peptone (Difco) water and aliquots (0.1 ml) were spread-plated in duplicate on Baird-Parker agar (Difco) for staphylococci and modified Oxford agar (Difco) for *Listeria*. Plates were incubated at 35°C for 48 hours before enumerating the colonies and calculating logarithmic values.

Non-microbial analyses

Thicknesses of nine pellicles, eight measurements each, were measured to the nearest 0.1 mm using an electronic digital caliper (Model Max-Cal, F.V. Fowler Co., Inc., Newton, Massachusetts). Slices of brined, air-dried salmon displaying the pellicle were photographed at 40× magnification through a dissecting microscope (Model SMZ-10, Nikon, Inc., Tokyo, Japan) by replacing one of the oculars with a digital camera (Model COOLPIX 5000, Nikon). Proximate analysis consisted of protein, fat, water, salt, and ash determinations. Protein, in 3 g of pooled samples in triplicate, was quantified on a nitrogen analyzer (Model FP-2000, LECO Corp., St. Joseph, Michigan). Lipid, in 24 g of pooled samples in duplicate, was quantified using an accelerated solvent extractor (Model ASE 200, Dionex Corp., Sunnyvale, California). Percent total solids was determined using method 952.08 (Cunnif 1996). Percent moisture, in 24 g of pooled samples in triplicate, was calculated directly from the gravimetric analysis of total solids. Percent salt was determined in triplicate using a chloride analyzer (Model 926, Nelson-Jameson, Inc., Marshfield, Wisconsin). Percent ash, in 6 g of pooled samples in triplicate, was determined using method 938.08 (Cunnif 1996). Water-phase salt (WPS) was calculated from salt and moisture data (Hildebrand 2001). Water activity (a_w), in 3 g of pooled samples in triplicate, was determined using a Model Aqua LAB Series 3TE instrument (Decagon Devices, Inc., Pullman, Washington).

Commercial samples

Pellicles from sockeye salmon (O. nerka) products being smoked were collected from three smokehouses (plants X, Y, and Z) in Alaska. Pellicles from plants X and Z were excised from hot-smoked salmon strips. Two batches of pellicles from plant Y were excised from pre-smoked salmon strips. Samples were air-shipped overnight to the FITC microbiology laboratory in small insulated boxes with ice-packs inside. The samples were analyzed for proximate composition, a_w and enumeration of aerobic plate counts (APC) and *S. aureus*, in addition to testing for the presence of *Listeria* species. Tests for APC, *Listeria* and *S. aureus* followed standard methods (USFDA 2001a). Colonies from APC plates were selected randomly (http://www.randomizer.org) and were

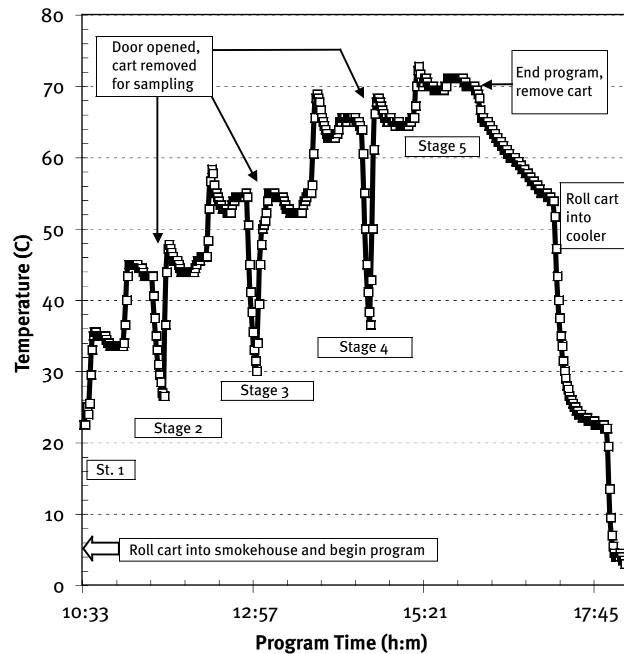


Figure 1. Temperature profile for a five-stage, six-hour smokehouse process for salmon strips.

Figure 2. Pellicle samples excised from hot-smoked salmon strips on racks removed from the smokehouse.

identified to *genus* and *species* by carbon substrate utilization patterns (MicroLog 2 version 4.2, Biolog Inc., Hayward, California). Presumptive-positive *Listeria* species and *S. aureus* were confirmed using the MicroLog 2 system.

Results and discussion

There are three phases of a typical hot-smoking program (Dillon et al. 1994): the fish are smoked for 0.5-1 hour at 30°C, often referred to as the tempering step, which promotes drying and toughening of the skin; followed by a heating step (50°C for 1 hour); and then a cooking step (70-90°C for 1-2 hours) and cooling to refrigeration temperatures before packaging, storage, and distribution. As an alternative to burning wood chips or sawdust for smoke generation, liquid smoke is applied during the first few hours of the process (Hildebrand 2001). The solubilization of smoke ingredients in water allows removal by sedimentation and filtering of the crude tar fraction containing potential carcinogens (Dillon et al. 1994). Liquid smoke can inactivate effectively *L. monocytogenes* (Vitt et al. 2001) and *S. aureus* (Paranjpye et al. 2004) on cold-smoked salmon.

Salmon strips that had undergone the smokehouse program resulted in a temperature profile verifying when samples were obtained (Fig. 1). About 20 min was required for removing the cart, obtaining the composite samples (Fig. 2), and re-installing the cart. Pellicle thicknesses averaged 1.3 ± 0.1 mm. A cross-section of a typical pellicle illustrated the ~1 mm thickness and underlayer (Fig. 3).

The composition of pellicles differed from the under-

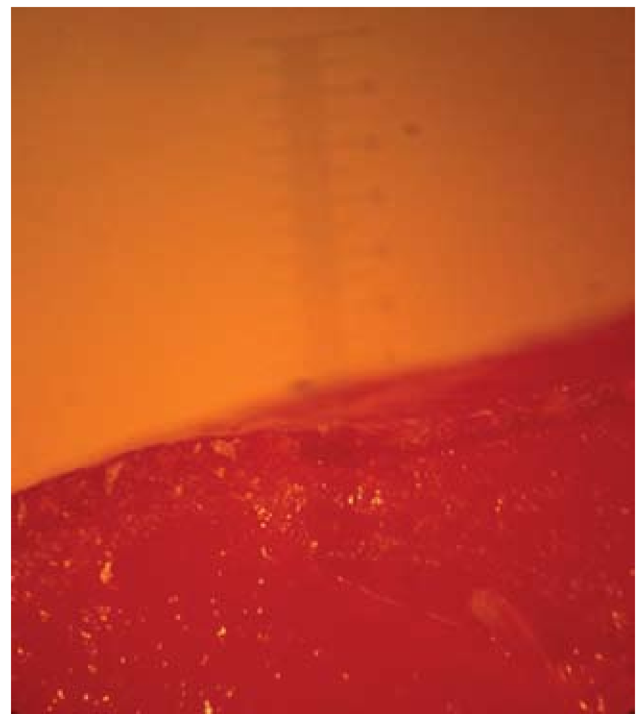
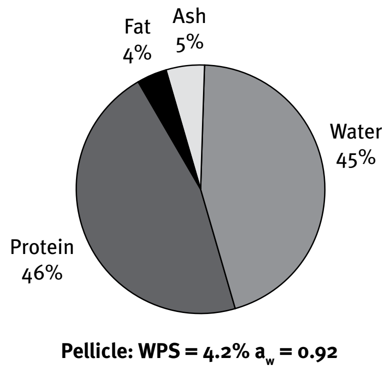
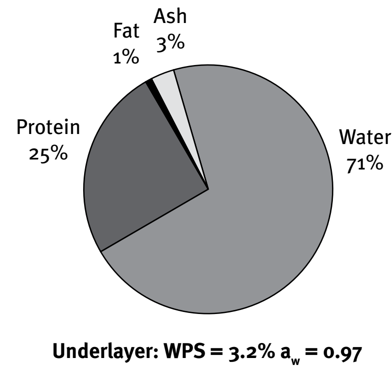


Figure 3. Cross-section of pellicle and underlayer photographed through a dissecting microscope (40× magnification).



Pellicle: WPS = 4.2% a_w = 0.92

Figure 4. Proximate analysis, water-phase salt, and water activity of salmon pellicles.



Underlayer: WPS = 3.2% a_w = 0.97

Figure 5. Proximate analysis, water-phase salt, and water activity of salmon underlayer.

layers with regard to percentages of protein, fat, moisture, salt, and ash (Figs. 4 and 5). Standard deviations about the means were between 0.2 and 0.7% for the proximate analysis of pellicles and underlayers. Pellicles were about twice as concentrated in the components as the underlayers due to dehydration. Water-phase salt was 1% higher for pellicles than for the underlayer (Figs. 4 and 5) with standard deviations of 0.38-0.57%. Water activity was 0.05 units lower for pellicles than for underlayers (Figs. 4 and 5) with a standard deviation of 0.005-0.006 units.

Commercial pellicle samples had lipid values twice as high in samples from plants X and Z (Fig. 6) and half as high in the two samples from plant Y when compared to the values for pellicles prepared in the FITC pilot plant. The variability between the proximate analyses of pellicles likely resulted from sampling at different stages of smoked salmon operations and different species of salmon being processed. Due to the limited amount of commercial pellicle samples available, water-phase salt was not determined. A mean a_w of 0.97 was recorded for the commercial samples. This high value is likely due to an equilibration occurring between the pellicle and adhering underlayer in the small sampling bags during overnight airline transportation to FITC. Bacterial content of the pellicles showed a range of 4.3-7.3 logs for APC (data not shown) and identities of randomly picked purified colonies showed a 2:1 prevalence of lactic acid bacteria (*Lactococcus lactis*, *L. garvieae*, and *Enterococcus malodoratus*) to gram-negative bacteria (*Shewanella putrefaciens*). Species of *Listeria* were detected in four of seven pellicles, although the MicroLog 2 system could not distinguish among *L. innocua*, *L. monocytogenes*, *L. seeligeri*, and *L. welshimeri*. The most probable number of *S. aureus* ranged from 93 to 1,100 per g in three of seven pellicles (data not shown).

Both inocula, *L. innocua* and *S. epidermidis*, showed a reduction of 4-4.5 logs in counts for the pellicle samples between post-brined dried strips and the hot-smoked products (Fig. 7). A control, absence of wood smoke, was conducted using the same inoculum levels and resulted in only 2.1-2.2 log reductions (Fig. 8). In comparison, *L. monocytogenes* can survive hot-smoking up to 68.3°C (155°F) when smoke is added throughout a 6-hour process, up to 82.2°C (180°F) when smoke is added only during the last half of the process, and up to 85°C (185°F) when no smoke is generated (Poysky et al. 1997).

An alternative method for applying smoke, in liquid form, was conducted after post-drying of the strips and inoculation with *L. innocua* and *S. epidermidis*. Liquid-smoked strips reduced the counts by 5.1 logs, while the controls (no liquid smoke) were reduced by only 2.3-3.1 logs (Fig. 9). In a third experiment, liquid smoke was added initially and compared with adding the smoke midway in the program. Inocula were reduced 4.1 logs when liquid smoke was added initially and 3.1-3.3 logs when added midway in the process versus a 2.2 log reduction in the control strips (Fig. 10). Liquid smoke, depending on the concentration applied and duration, is effective in reducing the maximum temperatures for surviving *L. monocytogenes* in hot-smoked salmon steaks (Poysky et al. 1997) and cold-smoked salmon (Vitt et al. 2001).

In hot smoking, the pellicle layer is dispersed by heat-coagulation of proteins and the drying step may be omitted (Autio et al. 2004). It is assumed that some drying does occur during racking, moving brined fish into the smokehouse, and delays prior to program start-up and heat input. The amount and rate of pellicle formation will be dependent on factors such as brine concentration, salt crystal size and quality, fish species, fish quality and freshness, protein and lipid composition, time, temperature, and the conditions of freezing and thawing of raw fish prior to smoke processing. Hot smoking can occur immediately after racking since the pellicle can be destroyed by heat, but cold smoking requires a certain amount of drying prior to smoking to help produce the pellicle (Dillon et al. 1994). A transition to a crust-like appearance occurs during the thermal treatment of dried pellicle. Too much drying initially in the smokehouse program can lead to case hardening (Dillon et al. 1994, Horner 1997, Doe et al. 1998, Hildebrand 2001, Eklund et al. 2004).

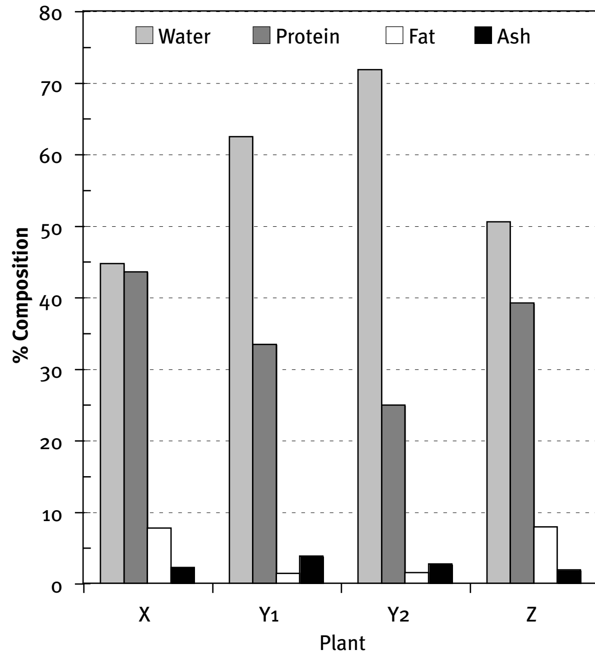


Figure 6. Proximate analysis of pellicle samples from three commercial smokehouses.

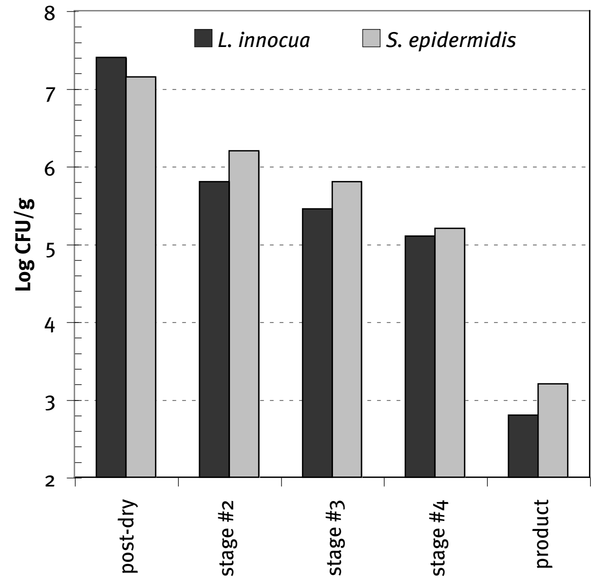


Figure 7. Reduction in *L. innocua* and *S. epidermidis* counts during hot-smoking of salmon strips.

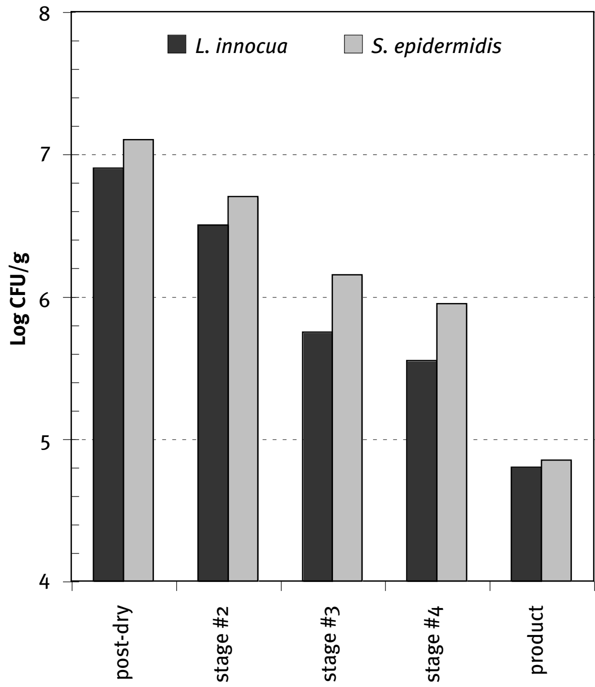


Figure 8. Reduction in *L. innocua* and *S. epidermidis* counts during hot-processing of salmon strips in the absence of wood smoke.

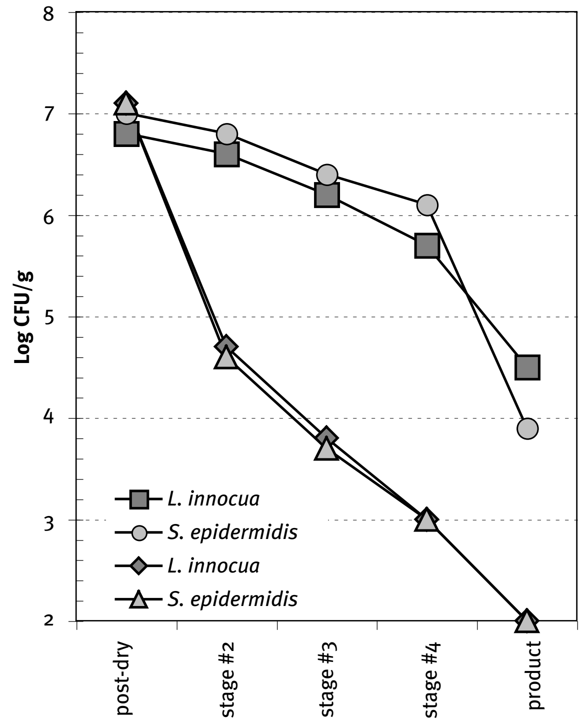


Figure 9. Reduction in *L. innocua* and *S. epidermidis* counts during hot-processing of salmon strips and in the presence of liquid smoke. Square and circle symbols refer to hot-processed (no smoke) salmon samples. Diamond and triangle symbols refer to liquid smoked salmon samples.

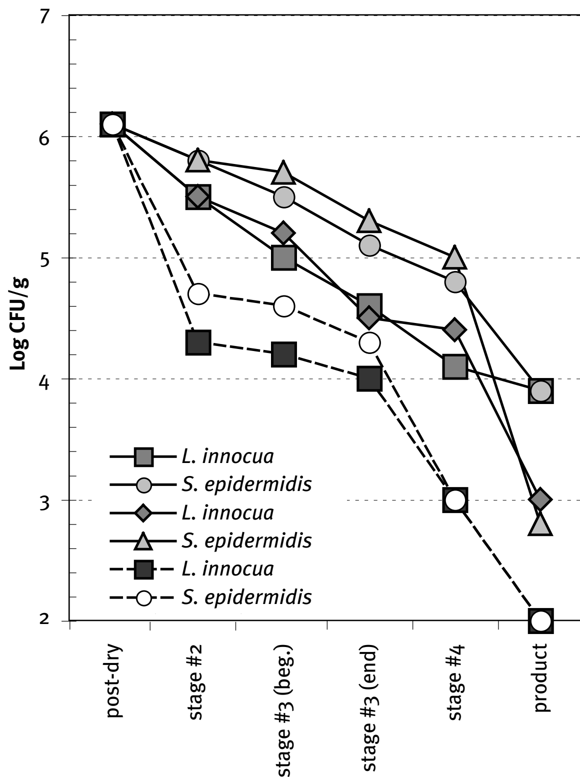


Figure 10. Reduction in *L. innocua* and *S. epidermidis* counts during hot-processing of salmon strips and in the presence of liquid smoke added initially or at stage 3. Square and circle gray-filled symbols refer to hot-processed (no smoke) salmon samples. Black squares and open circles refer to liquid-smoked salmon samples in which liquid smoke was added initially. Diamond and triangle symbols refer to liquid-smoked (at stage 3) salmon samples.

This study differed from all others in the field of smoked fish bacteriological research, since the focus was on sampling the pellicle layer (~1 mm) of inoculated salmon. Inhibition of bacterial contaminants at the surface of smoked fish muscle was presumed due to the combination of higher water-phase salt, lower a_w, higher smoke concentrations of antimicrobial constituents (i.e., phenols and acids), and higher temperature than in the underlayer. It is expected that *L. monocytogenes* and *S. aureus*, if present at low levels in salmon, would become undetectable regardless of the smoke type and when smoke was added during commercial processing.

Prevalence of *L. monocytogenes* occurs more often and in higher proportions for cold-smoked fish samples than for hot-smoked fish (Jinneman et al. 2007). Undoubtedly, the raw nature of cold-smoked fish retains viable bacterial cells that have entered the process at susceptible steps (Himelbloom 2008). However, the formation of a pellicle protects fish flesh (Autio et al. 2004), is essential for cold-smoked fish products (Dillon et al. 1994), and needs to be explored further with regard to safety of cold-smoked seafood.

Acknowledgments

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International Regulatory Requirements for Smoked Fish: Europe and Codex Alimentarius

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Part I. Europe

Introduction

The member states of the European Union (EU) are the largest importers of food worldwide. The EU is also the largest importer of seafood products. In 2005 about 9.9 billion pounds were imported into Europe with a per capita consumption rate of about 53 pounds. Europeans also are have a high consumption rate of smoked products including seafood. For smoked seafood producers in the United States, the high demand of the European market represents some genuine opportunities for selling their products. The EU does have significant barriers to importation of fishery products. The food laws are complex and are constantly changing and developing. Importers are subject to regulatory requirements imposed by both the European Commission and individual member states that are often confusing and conflicting. There are complex paper certification requirements and also numerous trade barriers. These barriers make it more difficult to do business in Europe and offset the potential opportunities driven by the demand for smoked fish products.

Overview of U.S. requirements for smoked fish

Smoked fish is subject to several food safety hazards. The most important one is *Clostridium botulinum* in products packaged in heavy film that limits product interface with air, which limits the ability of aerobic bacteria from growth and spoilage of the product. This packaging is known as modified atmosphere packaging (MAP) and it usually has gases such as nitrogen or carbon dioxide flushed into the package or a vacuum. In order to limit the probability of botulinum growth the water phase salt (WPS) level of the product should be at least 3.5% and the finished product should not exceed 38°F. Other requirements are (1) there should be 0 live parasites found in the product and (2) the *Salmonella* and *Listeria monocytogenes* levels should also be 0.

Overview of EU requirements for smoked fish (and ready-to-eat foods)

The European Commission presently has no specific regulations or guidelines for smoked fish and fishery products. Smoked fish is considered a ready-to-eat food. EU requirements for these commodities are found in Commission Regulation No. 2073/2005 on the Microbiological Criteria for Foodstuffs. Water phase salt for modified atmosphere packaging must be equal to or greater than 3.0%; there can be no live parasites; *Listeria monocytogenes* levels must be less than 100 colony forming units per gram and 0 for foods intended for infants or immune compromised persons; *Salmonella* must not exceed 0 CFU per 25 grams; coagulase positive *Staphylococcus* must be less than 100 per gram in cooked shellfish and crustaceans; *E. coli* must be less than 1 CFU per gram in cooked shellfish and crustaceans.

Commission Regulation No. 2073/2005 also requires that for ready-to-eat foods, several measures must be taken by food processors to control the growth of pathogens. These are (1) ready-to-eat foods must be monitored for *Listeria* as well as other possible pathogens; (2) food business operators are required to take measures to address "unsatisfactory results," i.e., unacceptable levels or presence of pathogens; and (3) food business operators are required to perform analysis of trends.

There are several other EU requirements that the food processor needs to know. For histamine-forming species the maximum allowable histamine level is 100 mg per kg, except for fermented processes where the limit is 400 mg per kg. The EU has limits for total volatile nitrogen and trimethylamines, but they are not specified in this document. The polycyclic aromatic hydrocarbon (which are present in most smoked products) limit is 5.0 mg per kg.

Parasite examinations for visible dead parasites must be conducted before the product is placed on the market. Cold smoked herring, mackerel, sprat, and wild Atlantic and Pacific salmon must be held frozen at –20°C for not less than 24 hours to control live parasites.

There are two other miscellaneous requirements. Food business operators must carry out organoleptic examination of fishery products to ensure compliance to EU freshness criteria, transport temperatures for chilled products should be near 0°C, and frozen products should be below –18°C.

Processors who cook (i.e., hot smoke) crustaceans and mollusks must ensure that the product undergoes rapid cooling following cooking. Water must be clean and the final temperature of the product must be near 0°C. After cooling the product must be chilled or frozen immediately. Additionally, shelling and shucking must be carried out hygienically.

EU certification

The European Commission requires that all shipments to the EU member states be accompanied by an EU Health Certificate. Furthermore, all shippers must appear on the EU Shippers List maintained by the Food and Drug Administration (FDA). Firms that wish to appear on the list must apply to FDA and be subject to a regulatory inspection that shows compliance with U.S. laws and regulations.

The present public health certificate will be merged with the animal health certificate effective November 1, 2007. For live and aquacultured products, animal health attestations will be required. For wild product the animal health section will be crossed out. In addition the new certificate will require the following.

- All packages shall bear "USA" and the approval/registration number (except bulk).
- Certificate list
    - ISO code.
    - Harmonized commodity code.
    - Identification of the container/seal number.
    - Flight/voyage number.

The inspector will attest to the following as true and accurate:

- Public health—the firm is/has
    - Compliant to U.S. Code of Federal Regulation as prescribed in Counsel Decision 98/258/EC.
    - Adequate health control systems in place (e.g., HACCP).
- Animal health—aquacultured products:
    - Certain species do not have certain diseases (e.g., chinook salmon do not have VHS).
    - That certain species do not come from certain areas or regions.

Two other regulatory requirements that firms doing business in the EU should be aware of are labeling requirements and traceability. The EC has complex directives that cover labeling that should be followed. Also each member state may have specific labeling requirements, and country of origin labeling is required everywhere in Europe. Europe has so-called traceability directives that require extensive records to be in compliance. It is much more comprehensive than the "one forward, one back" system of product tracing utilized in the United States.

Part I conclusion

European standards for smoked fish appear to be less stringent that the United States and Canada. However, exporters to the European Union can expect to encounter problems with certification, microbiological testing, and chemical testing of shipments. Also, label requirements will vary by member states, and Europeans have significantly different taste preferences for smoked fish from North American consumers.

Part II. Codex Alimentarius—What is it and why does it matter?

Codex Alimentarius is the primary international food standards body, sponsored by the World Health Organization (WHO) and the Food and Agriculture Organization (FAO). It has many so-called vertical committees that cover standards for most food commodities as well as so-called horizontal committees that cover cross-cutting issues such as food hygiene, labeling, and analytical methodologies. Commodity committees such as the Codex Committee for Fish and Fishery Products (CCFFP) generate standards and guidelines that relate to a particular commodity. Standards for a certain product and Codes of Practice are the most common documents that are generated by these committees. A standard sets the minimum criteria for food safety for a certain commodity and a Code of Practice gives advice about how to attain the standard through manufacturing best practices.

Codex Alimentarius began in the 1960s and functioned primarily as a source of information for developing countries. When the World Trade Organization was established, it adopted Codex Standards as a basis for evaluating sanitary and phytosanitary measures in trade. Countries are often accused of creating food safety standards that are not science-based but designed to reduce trade in a given commodity. Since Codex Alimentarius standards are science-based and derived by broad international consensus, they became the ideal basis for determining if a food safety measure was a trade barrier. This caused Codex Alimentarius delegates to be extremely careful in ensuring that their country's best trade interests were considered when developing the standards.

Both the Codex Alimentarius standard and Code of Practice for Smoked Fish are currently before the CCFFP for development. The main issues are the minimum level for water phase salt (WPS) that will ensure food safety in modified atmosphere packages and whether the standard should allow the use of liquid smoke. The North American view of WPS is that vacuum or modified atmosphere packages need to have a WPS level of 3.5% while most Europeans believe that 3.0% is sufficient in order to prevent the possible growth of *Clostridium botulinum*. The science on *C. botulinum* growth in salty media is not sufficient to definitively determine the exact level of WPS necessary. The United States cannot accept a WPS level of less than 3.5% because we believe that it would not sufficiently protect the U.S. consumer. Europeans prefer less salty tasting smoked fish and are adamant that extra salt is unnecessary. It is likely that the committee will deadlock and no international standard will be established. The U.S. delegation has offered a compromise approach we term "the matrix" which allows each country to set their own level of WPS that they believe will protect their consumers.

Here is the proposed compromise:

Storage temp.	Packaging	Water phase salt	Comments
0°C to 3°C	Any	No minimum	Temperature monitoring on each package.
>3°C to 5°C	Aerobic packaging	No minimum	Storage temp is for general pathogen control and quality. Sensory signs indicate spoilage.
Frozen (≤–18°C)	Reduced O_2 or VAC/MAP	No minimum	*C. bot.* cannot form—need label information when thawed.
>5°C to 5°C	Reduced O_2 or VAC/MAP	3.0 to 3.5% selected by country where product is consumed	WPS at least 3.0-3.5% will delay/prevent toxin formation.
>5°C to 10°C	Reduced O_2 or VAC/MAP	5%	Non-proteolytics (*C. bot.*) controlled.

C. bot. = *Clostridium botulinum*. WPS = water phase salt.

This proposal will hopefully be accepted by the committee when it meets in Trondheim, Norway, in February 2008 and will result in a developed international standard for smoked fish.

The other important issue concerning smoked fish before the CCFFP is the use of liquid smoke. The use of this additive brings up several questions, e.g.: Is a product treated with liquid smoke actually smoked? Is liquid smoke used primarily as a food additive or as a source of smoke generation? What about when liquid smoke is used to enhance flavor in a traditional smoking process? The European delegations objected to liquid smoke inclusion in the smoked fish standard so the committee is currently deadlocked. The U.S. delegation will advocate that liquid smoke be included in the standard and if used should be properly labeled as a food additive. It is noteworthy that the European Commission has a Directive on Liquid Smoke (88/388/EEC) so it is obviously used as a food additive in Europe.

Part II conclusion

Codex Alimentarius issues are important because they may affect a food producer's ability to trade in fish and fishery products. The only way the CCFFP can succeed in developing an international standard for smoked fish and ensuring fair trade is through compromise on the issues of water phase salt and liquid smoke. The U.S. delegation will work hard to produce acceptable results for both the Smoked Fish Standard and the Smoked Fish Code of Practice.

HACCP Plan

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The Hazard Analysis and Critical Control Point (HACCP) plan and sanitation monitoring records are the main documents required by part 123 of the Code of Federal Regulations. Most industry members use a HACCP plan format (see HACCP Plan Form) similar to the one offered by the Hazards and Controls Guide (USFDA 2001, 3rd edn.). A HACCP plan is the document that summarizes the industry member's knowledge of the hazards involved, if any, and their plan to control those hazards. The plan also helps the inspector judge the industry's ability to produce a safe and wholesome product.

This presentation covers observations from FDA inspections since 1997 and provides suggestions to correct and improve an existing HACCP plan.

Critical control points

In the earlier days of HACCP, a few critical control points (CCPs) were typically found to be omitted from HACCP plans, particularly the refrigerated storage and the brining steps. Today, most CCPs are correctly included.

A trend for many processors in the past couple of years is to label and ship their product as frozen. Choosing to label, store, and ship their finished product as "Keep Frozen" eliminates the involved CCPs for *Clostridium botulinum* control, but may possibly lead to a labeling CCP depending on the likelihood for their product to be mislabeled. The complete "Keep Frozen" statement recommended by FDA is in the Hazards and Controls Guide in Chapter 13. Many of these "Keep Frozen"–type HACCP plans have only one CCP—parasites—unless the product labeling is also determined to be a CCP.

The most commonly stated "significant hazard" error is only listing "pathogens," which doesn't reveal which pathogens the critical limit is targeting. It is well accepted that *Listeria monocytogenes* and *Clostridium botulinum* are the target pathogens for which HACCP plans are designed. So stating those two pathogens in your HACCP plan eliminates any doubt what specific hazards the critical limits are designed to control.

Additionally, parasites would be a significant hazard required for a cold smoked product and can be controlled prior to receipt or by freezing at some step like receiving, storage, or finished product storage. Parasites are also a significant hazard for a hot smoked product, and are most typically and easily controlled at the smoking step.

Critical limits

The most common error in "critical limits" for smoked products used to be indicating "3.5% water phase salt" (WPS) as a critical limit. Nobody was actually measuring the water phase salt of each lot, yet that is what was listed as the critical limit to control. Now nearly everyone recognizes the need to list the things they actually control as critical limits, in order to attain the correct WPS.

HACCP plans for smoked products are probably the most difficult to write because there are several CCPs and critical limits working in unison to control all the hazards.

Monitoring

The "What" and "How" monitoring in the HACCP plan is generally straightforward and only a few errors were noted in these two areas. The internal temperature is monitored in hot smoked products with the smoker's temperature probe because it is necessary to ensure all parts of the smoked fish reach the critical limit. Nearly everyone was doing this correctly, which involved ensuring the probe was placed in the thickest portion in the coldest part of the smoker. However, some processors were also placing probes into the center of their cold smoked product, which does not reveal useful information.

The goal in cold smoking is to prevent the reduction of spoilage organisms, particularly on the surface of the product. Placing a probe in the center of the product will show a much cooler temperature than the smoking cabinet air temperature. It is important to closely monitor the smoking cabinet air temperature so the spoilage bacteria are not killed. In the event of subsequent time/temperature abuse we are relying on the spoiled appearance and odors to alert the consumer that this product is not fit for consumption. If the consumer can't get the product past their nose then they will not consume a product that may also have produced *Clostridium botulinum* toxin during the time/temperature abuse.

This spoilage and water phase salt content are the sole barriers for controlling the pathogen and *Clostridium botulinum* toxin growth in the refrigerated finished product.

Knowing the hot or cold spots in your smoker is essential to producing a safe product. Documenting the warmest/coolest part of the smoking oven will be covered in the verification section.

There was initially confusion about the correct interpretation of "continuous" monitoring in the "Frequency" section

HACCP Plan Form

Firm Name: Firm Address:	Product Description: Method of Storage and Distribution: Intended Use and Consumer:

(1) Critical Control Point	(2) Significant Hazards	(3) Critical Limits for each Preventive Measure	Monitoring				(8) Corrective Actions	(9) Verification	(10) Records
			(4) What	(5) How	(6) Frequency	(7) Who			

Signature of Company Official:	Date:

of the HACCP plan. The current commonly accepted method typically includes the use of recorder charts or data loggers. Now the most common mistake in the frequency section is leaving out the check against a known accurate thermometer at least once per batch.

In the "Who" section of the HACCP plan there are rarely any errors or confusion about who needs to perform monitoring.

Corrective actions

Although listing "corrective actions" is optional in any HACCP plan, most industry members will include their preplanned specific directions for corrective actions, which is helpful to plant personnel. Preplanned corrective actions must address two parts:

1. Ensuring no suspect product enters the market; and
2. Ensuring that the cause of the process deviation is corrected.

In the beginning we observed that at least one of these criteria was missing from most firm's preplanned corrective actions. Now, nearly everyone understands their preplanned corrective actions need to address the two items mentioned above.

Records and verification

Past problems in recordkeeping and verification parts of the HACCP plan include "not reviewing records within seven days as required, not specifying an appropriate periodic interval for calibration or verification activities and not recording calibrations that do take place."

In Alaska, scales are often overlooked as needing calibration because a state agency performs annual scale inspections and applies an annual certification sticker to each scale. Scales used to monitor the amount of fish, salt, sugar, brine, water, etc. must be periodically calibrated to ensure the critical limits are being met.

It's important to consider what calibrations are necessary for all the CCP monitoring equipment. For most critical limits an instrument such as a thermometer, timer/clock, or scale is used to measure critical limits. Such equipment must be periodically calibrated to ensure it can measure the critical limit accurately. This was not being done in the initial HACCP plans. Now nearly everyone understands the need for calibration.

Hot and cold spots in the smoking oven need to be verified, or at least the lack of a hot or cold spot needs to be verified. Many times the manufacturer can provide verification or certification that the temperature in their model of smoker doesn't vary significantly. Home-built smoking ovens, or those ovens that have been modified after purchase from the original manufacturer, will need additional assurances. A heat distribution study would have to be conducted to confirm there are no hot or cold spots in the smoker. It is of little use to have a critical limit that isn't applied uniformly throughout your smoker. The verification documents showing that the smoker distributes heat evenly should be included as a verification record and made available to the inspector. If the smoker doesn't heat evenly we would want to know where the hot/cold spots are to ensure they are where monitoring occurs during the smoking/heating step.

Sanitation plans

The state of Alaska requires a written sanitation plan, while the federal regulation does not. Initially, a common omission in many firms' sanitation monitoring records was any of the eight sanitation points relevant to the plant. Now, the most common sanitation monitoring observation is plant personnel noting a sanitation problem, but omitting the corrective action or the date and time of the correction. It is important to not only document the problem, but to document the corrective action.

It's also a common sanitation monitoring problem to omit the name or initials of the person who made the observation. Also, when checking chlorine levels in processing waters, using a check mark instead of an actual parts per million (ppm) measurement is inadequate. When sanitation issues are properly documented the firm is able to track trends.

Definitions needing explanation

Continuous monitoring: Recorder charts are straightforward continuous monitoring instruments for most people, but data loggers can be quite variable. On a large freezer with little variation in temperature, the interval between recorded measurements may be large. But on a small smoking oven with blowers, changes can occur rapidly so the interval between measurements must be very small.

The proper interval for any application will be one insuring no critical limit could possibly be exceeded. The interval will depend on how fast the temperature can change and how close the operating limits are to the critical limits. Crab cookers with hundreds of gallons of boiling water which typically operate 30-35°F above the critical limit can realistically have longer intervals. A smoker that has a fan forcing smoke and heat throughout the smoker cabinet typically operates at only 5-10°F above the critical limit for about 30 minutes. Should the heat fail on a smoker, the fan can quickly cool the product, allowing the critical limit to be missed.

Periodic monitoring: This is most important when used in conjunction with calibration. Much like the "continuous" statement, periodic means, "often enough to ensure the calibration can be assured." When using dial thermometers, calibration may be daily. Thermocouples or radio tracking devices or similar equipment may be calibrated on a monthly or longer schedule. Typically, the "known accurate thermometers" used to calibrate all the others within the plant have a high degree of accuracy and are readable to one degree or less and are typically calibrated annually.

If the FDA checks the accuracy of thermometers and finds them to be off by several degrees, we may require a review of the calibration methods. If the methods are acceptable the periodic interval for calibration will be questioned. A more definitive explanation for periodic calibration intervals, forthcoming in the 4th edition of the Hazards and Controls Guide, may better define "periodic" and eliminate confusion.

Training: HACCP training is required to develop and reassess HACCP plans and to review monitoring records. "Training" can be going through a standardized curriculum or can mean that experience has resulted in an equivalent education.

Interpretation of these somewhat vague regulations falls to the inspector in the field. Generally, the inspector will judge the level of competency based on the HACCP plan and available records. A decent HACCP plan combined with good CCP and sanitation monitoring records, along with demonstrated knowledge, will indicate that the equivalence standard has been met.

HACCP training isn't required for reviewing monitoring records, but demonstrated adequate knowledge of HACCP is.

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Compliance with FDA's HACCP Regulation

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Abstract

A smoked seafood processor must comply with many local, state, and federal regulations, including the Seafood Hazard Analysis Critical Control Point (HACCP) regulation. In brief, the regulation requires training, development, and implementation of a HACCP program, and monitoring of eight designated sanitation elements.

Although it may appear to be a difficult task for a new processor to fulfill the requirements of the regulation, the processor who spends the time and resources to develop a HACCP program that complies with the regulation will save considerable resources in the long run. Failure to comply can result in state and/or federal regulatory action that can overwhelm a processor financially, and affect their reputation in the market place.

This presentation is intended to give honest and open insights on critical elements of the regulation, and suggestions on how to comply and avoid regulatory action from the perspective of a former manager of the FDA and current industry consultant. The presentation is based on the experience of reviewing HACCP program for virtually all fish and fishery products, and case development that resulted in regulatory actions, including injunctions.

Introduction

I had a 32 year career with the U.S. Food and Drug Administration (FDA) before I retired in January 2005. At FDA, I managed the seafood inspection and sampling programs including the Seafood HACCP Regulation Enforcement Program.

During my tenure, I reviewed hundreds of HACCP programs—including those for many smoked seafood operations—and initiated regulatory action when voluntary compliance was not achieved. Since then, as an independent consultant, I have also audited HACCP programs for several different commodities including smoked seafood products. Therefore, I have reviewed and evaluated many HACCP programs from both sides.

I want to give you the perspective of an FDA regulator and as an independent consultant on several important aspects of the HACCP regulation. Obviously, I can't cover everything but will focus on some major problems I saw as a regulator and now see as a consultant.

My talk is geared toward the individual getting into the business, but hopefully it will also be beneficial to those currently operating smoked seafood operations.

Objectives

First, it should be acknowledged that the primary goal of everyone in attendance today is the production of safe and wholesome products—be it industry, regulatory, academia, industry association, or consultant. We all just have different responsibilities in achieving the goal.

For a new or established processor, one of your major concerns has to be compliance with the various local, state, and federal regulations, and it's important you know the subtle differences between the different agencies and their requirements. For example, both Alaska Department of Environmental Conservation (ADEC) and U.S. Food and Drug Administration (FDA) enforce the HACCP regulation, but there are subtle differences in regard to the hazard analysis, finished product storage temperature requirements, and sanitation standard operating procedures. If you operate in other states across the country, there may also be similar differences. So you must take the time to learn the basic regulation and the subtle differences between the various agencies. However, if you are involved in interstate commerce, you must comply with the FDA's HACCP regulation, at a minimum. States have the option to exceed the minimum requirements of the FDA regulation.

Depending upon your customers, you may also have to comply with their additional requirements. For example, if you want to sell product to the military, they have additional requirements that are not included in the FDA regulation, and probably not in the state regulation. If you want to sell to the "big box" stores—such as Costco or Wal-Mart—you will be expected to meet their higher standards above and beyond the mandatory regulations. These customers are extremely concerned with product liability, as you should be, and will have your processing plant inspected by third party auditors that evaluate compliance with the regulation and other designated program requirements.

Hopefully, despite the regulatory and customer requirements, you can still make a product that satisfies the customer and yields a reasonable profit.

[1]U.S. Food and Drug Administration, retired.

Training

Seafood HACCP training is a basic requirement of the regulation that must be met by you and/or your staff, unless you contract with a HACCP trained person to perform specific designated functions. The training must be one of your first priorities, and it will give you information on the basic HACCP principles and terminology.

However, despite the gigantic effort by the regulators, academia, industry associations, and all the other trainers, the reality is the training has limitations and should be the first step of several steps in understanding and implementing HACCP. Based on conversations with many processors—from both perspectives—the training does not provide adequate knowledge and tools necessary to develop a compliant HACCP program. This is especially true for small companies. This doesn't mean the curriculum is bad or the trainers are not doing a good job. Rather, the real problem is that individuals come to the training course thinking all they have to do is attend a training course and they are totally prepared, and they will have the opportunity to develop a HACCP program during the course for their specific plant and products. The HACCP training format simply doesn't allow for that approach. As a result, some individuals become discouraged and do not follow through after completion of the training course.

Unfortunately, some individuals take the training, get their certificate, put the training manual on the shelf, and rarely look at it again. Others procrastinate, make a limited attempt to comply, or look for shortcuts. In some cases firms have simply made a copy of another processor's HACCP program or used an Internet site to download HACCP programs, which they use as is.

To truly understand HACCP and be prepared to develop a compliant program, you must be prepared to go beyond attending a basic training course. You need to use the training materials, and seek out additional training and other sources of information to develop a HACCP program that is specific to your plant and products. There are several good sources such as the University of California Davis and other Sea Grant affiliates, the Alaska Sea Grant Marine Advisory Program, the Alaska Seafood Marketing Institute, and industry associations such as the GMA/Food Producers Association.

Another option is to use a qualified HACCP consultant to develop your program, especially if you're not willing to use the training received or put effort into the process of developing a compliant program. It may cost a little money up-front, but it could save you the tremendous costs involved if regulatory action is taken by FDA.

HACCP plan

Whichever approach you take, it's important that you develop a HACCP program that is specific to your plant and products. That's why you're cautioned about making copies or downloading HACCP programs and using them as is. They may seem to have the basic HACCP elements, but there are differences between plants, products, procedures, and equipment.

A key element of the HACCP program is establishing the critical limits for your process, especially the brining step. The verification section of your HACCP program must cite the validation study that established the critical limits. It's highly recommend that you contact the various sources—industry associations, academia, etc.—to have your process validated. This should ensure your process will always result in 3.0% or 3.5% water phase salt on a daily basis. This is a very common problem and has resulted in regulatory actions.

Another key element is implementation of the HACCP program you developed. A good HACCP program is no good if not implemented properly and completely. The FDA inspector will make an evaluation of the HACCP program developed, and whether it has been implemented properly. Essentially, the inspector will evaluate whether you are doing what you said you were going to do in the HACCP program. For example, if you say you are going to use a temperature recording device (versus a dial or digital thermometer) to monitor a refrigerator temperature, then the inspector will determine whether you are using a temperature recording device; if you say you will visually monitor temperatures every 2 hours, the inspector will evaluate whether this is being accomplished. Be sure to implement all aspects of your HACCP program.

If you are not following your HACCP program, it may mean you need to re-assess the procedures, equipment, etc., to reflect what you are actually doing on a daily basis. Be sure to follow the steps necessary to re-assess your program.

Organization attributes are not a requirement of the regulation but can be an important factor on how the inspector looks at your HACCP program. A well organized program creates a positive mind-set of your firm and HACCP program, and takes considerably less time for the inspector to review the program. You can usually expedite completion of the inspection if you have a well organized program.

Records

Another key element of a HACCP program is records that are logically designed for monitoring the critical limits. Many times records are formatted by a corporate official who has never been in the plant, let alone filled out the records. While the record may look good on paper, the record format may not meet the needs of the line person completing the record on a daily basis.

It is recommended that you don't create monitoring records in a vacuum. Involve the individuals who will be using the records every day to ensure their usefulness and compliance with the regulation. The verification review of records should identify this type of problem and the record format revised as necessary.

But most important, the records associated with monitoring critical limits at critical control points must be

completed in full and in real time. Blanks or the lack of entries on a record are red flags to the inspector. So are records with the same data entries day after day. For example, it's statistically impossible for a refrigerator to read exactly 38.0°F 365 days a year—but this pattern has been observed on many occasions.

Under no circumstances should you or your employees falsify records, or give the impression that records are being falsified. Record falsification is a Title 18 charge, a far more serious violation than noncompliance with the HACCP regulation. Never allow records to be pre-filled, pre-dated, etc. For example, a record with data for March 5 through March 9, but signed and dated as reviewed on March 7 raises a question about the integrity of the record and record keeping system. It's better to have an incomplete record, or no record at all, than a falsified record.

Records must be reviewed by a HACCP trained individual within designated time periods. Too often, this is a passive process and errors are not caught. Verification record review is a serious responsibility and a dedicated effort must be made during the process.

Several other aspects of good HACCP programs for smoked seafood products have already been discussed by other presenters and will not be repeated.

HACCP inspections

You should not consider the inspection an adversarial situation. Don't be paranoid about the inspector or inspection process. As they say, they are there to help you. If objectionable conditions are identified during the inspection, make sure you understand the issues and ask any questions you may have, to clarify the conditions discussed. Unfortunately, the processor's mind is usually going in a thousand different directions and is not focused on the discussion. As a result, the processor doesn't fully understand the issues and what needs to be done to correct the objectionable conditions and they don't always get corrected.

You should be cooperative and honest with the inspector. Creating a hostile environment will not be productive. In today's world, idle threats are not taken lightly by the agency. If necessary, the FBI will be contacted to intervene in these situations. This is the exception, but it does happen from time to time.

At the same time, you should be aware of your rights on some inspection procedures:

Plant Tour: Establish your policy, but it's recommended that you never allow anyone to roam your plant alone. With the inspector or a customer representative, someone should accompany the individual and be available to answer questions; with the general public, you should be concerned with food security issues and have someone accompany the individual.

Photographs: Establish your policy and follow it. If you decide no photographs, then make it clear from the very beginning that no cameras are allowed in the plant, period! Not just no photographs, but no cameras; there is a difference. Despite the reason typically given, the FDA does not have a specific court case giving them the authority to take photographs. Stay compliant and don't create a situation that could become a test case. Once photographs are taken, the film becomes the property of the government and will not be returned or destroyed.

Affidavits: You are advised to consult your attorney before signing any legal documents.

Sampling: You must allow the inspector to collect samples, but it's suggested you take duplicate samples. However, keep in mind the FDA analysis will be used no matter what your analysis reveals.

Written Request: It's recommended you give the FDA inspector a written request for a copy of their written narrative establishment inspection report. You are entitled to this under Federal Management Directive No. 145, but the agency does not routinely provide a copy unless a written request is made. Having a copy of the report will allow you to review their evaluation of your HACCP program and plant, and perhaps assure you they correctly interpreted your programs, procedures, etc.

Inspection results

The FDA uses the form FDA 483, Inspectional Observations, to identify objectionable conditions identified during the inspection. At this point you may be near a nervous breakdown, but as indicated before, take the time to read the observations, make sure you completely understand the observations, and ask questions so you fully understand the issue, possible solutions, and ramifications if you don't correct the conditions.

Next, correct the objectionable conditions as soon as possible. Some may be easy to correct, and some may take more time and resources to correct. Start the process during or immediately after the inspection. When an inspector (agency) sees the same problems year after year, it gives a negative impression of your firm. The agency relies upon voluntary compliance, and it's in your best interest to make the corrections.

After the conditions have been corrected, it's recommended you prepare a written response to FDA as soon as possible. Describe what you have done to correct each objectionable condition and provide records, photographs, and other documentation as necessary to verify the corrective actions taken in response to the inspectional findings.

If necessary, request a meeting with the local FDA management to review the correction actions in response to the inspection. Don't be afraid to interact with the agency. A meeting will give you a chance to meet and establish a dialog with FDA personnel, and a meeting may give you more direct feedback on your corrective actions. Sometimes, a meeting is more productive than exchanging letters with the agency.

Consequences

If objectionable conditions are found during the inspection, you may receive an untitled letter at some point after the inspection. The untitled letter is the proverbial "first bite of the apple." After the inspection, if the agency has not received a written response from you or does not consider your response adequate, they may send you an untitled letter. As the name implies, the letter does not have a title. It essentially summarizes what the inspector found during the inspection and asks for corrective action. It is not considered a regulatory action and it stays internal to the FDA.

Depending upon available resources or seasonality of your operation, FDA will make a follow-up inspection. If the inspection finds the objectionable conditions have not been adequately corrected, a warning letter may be issued. This is a significant letter, and it has a title, i.e., "warning letter." It summarizes the violations and requests a response within 15 days detailing the corrective actions taken, and warns that additional regulatory action may be pursued if the objectionable conditions are not corrected. If you get a warning letter, you are advised to respond immediately. In addition, warning letters get published on the FDA Web site and become available to anyone who conducts an Internet search of your company name.

FDA will make a timely follow-up to a warning letter. If significant deficiencies remain, they may pursue an injunctive action. This is an extremely serious situation and you will be expending resources for attorneys, laboratories, possible product recalls, and consultants. At this point, you will definitely wish you had put more effort into training, and developing and implementing a valid HACCP program, and had made corrections on a voluntary basis.

Common reasons for problems

As a manager with FDA, I confirm that the review of establishment inspection reports written by inspectors found many of the problems discussed today. Sometimes the same problems were observed inspection after inspection and it was a mystery why the conditions weren't simply corrected to avoid possible regulatory action. The problems appeared to be relatively basic and with a few dedicated hours correcting the objectionable conditions would resolve the problem once and for all. But that was not always the case. Although a "mystery," it was assumed that money, time, and priority were the root factors.

As a consultant to clients who have minor and serious compliance problems, especially those currently under injunction, I have confirmed some of the assumed reasons.

Money: Although some processors are willing to spend money on certain things, they become frugal when it comes time to spend resources for sufficient training, equipment, and employees necessary to comply with the regulation. Unless you are prepared to spend the necessary resources in these and other critical areas, you will have problems down the road.

Time: Employees responsible for HACCP are oftentimes required to perform too many other tasks, and HACCP may become a low priority as a result. Unless you make time available to employees to implement the HACCP program in your operation, problems will eventually develop. This can happen even if you start with a compliant HACCP program.

Priority: In some cases, the firm produces the product first and then tries to develop a HACCP plan after the fact. Obviously, this approach can lead to serious problems, especially if there is no follow-through and no plan gets developed.

In conclusion, by not expending the necessary resources and giving priority to HACCP when the problems were initially identified during inspections, these companies have had to spend extensive resources, have lost business, and most important they have lost their reputation.

You are encouraged to consider these issues to avoid problems with the regulatory agencies.

Conclusions

Despite the regulatory requirements, you can make a good product and a profit.

Process Establishment or Validation for Refrigerated, Vacuum-Packaged Hot-Smoked Fish, and HACCP Monitoring Considerations

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Vacuum-packaged, hot-smoked fishery products are a concern of FDA since pathogens may survive the hot-smoked fish process, and then cause illness after being consumed as a ready-to-eat (RTE) product. For this reason, FDA expects the adequacy of the hot-smoke process to be established and/or verified by scientific study, in order to demonstrate that the pathogen of concern is adequately destroyed, reduced, or inhibited to levels that are safe. The problem for processors in this regard is twofold. FDA Guidance (FDA 2001) provides very little insight into how to systematically design and conduct a scientific study necessary to establish and/or verify the adequacy of a process for smoked fishery products. In addition, while at the same time assuring product safety, processors are also concerned with optimizing process efficiency and product quality.

Process establishment/validation involves finding answers to four basic questions:

1. What is the pathogen of concern in a RTE smoked fish product (i.e., the target pathogen)?
2. What must the process deliver, in terms of final product attributes, in order to be considered safe?
3. Exactly how does one determine whether the smoked fish process is actually delivering to the final product those attributes necessary for adequately controlling the target pathogen?
4. What HACCP controls (critical limits, monitoring procedures, etc.) should be in place to ensure the process consistently delivers the final product attributes necessary for adequately controlling the target pathogen?

Concerning question 1, the target pathogen for control is dictated by the packaging style, and the method of finished product storage and distribution. For a refrigerated, vacuum-packaged (reduced oxygen packaged) smoked fish product, the target pathogen of concern is *Clostridium botulinum* type E and non-proteolytic type B and F. There are two different groups of *C. botulinum*, based on their physiological characteristics. These characteristics are summarized in the table below.

Addressing question 2, the process must assure that final product achieves two critical attributes for controlling non-proteolytic *C. botulinum*: both (1) a 3.5% WPS in the fish flesh, or a 3.0% WPS + 100 ppm nitrite in the fish flesh, *and* (2) a minimum internal core fish temperature of 145°F for minimum hold time of 30 minutes (WPS = water phase salt). There are other potential stand-alone controls for non-proteolytic *C. botulinum*, one of which is frequently achieved in hot-smoked fishery products (but seldom in cold smoked product); that being a water activity (a_w) of <0.97 in the fish flesh. Because of this, it is highly recommended that processors monitor the a_w in addition to percent WPS. Non-proteolytic *C. botulinum* can also be adequately controlled by achieving a WPS >5%, but this is not commonly achieved in hot- or cold-smoked fishery products.

Concerning question 3, conducting a scientific study to establish and/or validate the adequacy of the hot-smoked fish process is the method processors should use to determine whether the smoked fish process is actually delivering to the final product those attributes necessary for adequately controlling the target pathogen, non-proteolytic *C. botulinum.* In this regard, this scientific study must demonstrate that:

1. The *curing process* delivers the proper inhibitory level (concentration) of salt and/or nitrite to each and every unit of product (or reduces the a_w to a proper inhibitory level), and

C. botulinum type E and non-proteolytic B and F	*C. botulinum* type A and proteolytic B and F
Anaerobic	Anaerobic
Can grow/produce toxin at >38°F	Can grow/produce toxin at >50°F
Min. a_w = 0.97	Min. a_w = 0.935
Max. WPS = 5%	Max. WPS = 10%
Spoilage may not be evident (no odor, no gas)	Spoilage usually evident (obnoxious odors, gas)
Kill or damage to control	Refrigerate to control

2. The *hot smoking/drying process* delivers the proper internal product temperature for a specific period of time to each and every unit of product.

In the end, it is the complex interplay of smoke, salt, temperature, and nitrites that act together as an effective barrier to non-proteolytic *C. botulinum* growth and toxin formation.

Finally, in addressing question 4, the HACCP controls (critical limits, monitoring procedures, etc.) that should be in place to ensure the process consistently delivers the final product attributes necessary for adequately controlling the target pathogen will ultimately be determined by considering the process operating parameters under which the scientific study was conducted.

As previously mentioned, since there is very little FDA guidance regarding how to conduct a process validation study for refrigerated, vacuum-packaged hot-smoked fish products, the objective now is to examine the steps necessary to systematically conduct such a study. For any process validation work involving refrigerated, vacuum-packaged hot-smoked fish, one needs to remember the bottom line—providing a demonstration that the process (1) delivers the proper inhibitory concentrations of salt and/or nitrite to each and every unit of product, and (2) delivers the proper internal product temperature for a specific period of time to each and every unit of product.

There are four basic requirements involved in a process validation study:

Step 1 Designing the study that considers all factors and/or conditions that may affect attainment of the necessary finished product attributes.

Step 2 Gathering process and product attribute data, under a specific set of processing conditions.

Step 3 Analyzing data to determine if process study conditions result in achieving the finished product attributes needed for control of non-proteolytic *C. botulinum*.

Step 4 Identifying critical factors and/or conditions that could affect the adequacy of process, and establishing appropriate HACCP controls (critical limits [CLs], monitoring, records, etc.)

Each of these steps are examined here and discussed in more detail.

In step 1, study design, it is important to be knowledgable in your equipment design, function, and capabilities. Various processing conditions and system considerations include brine strength, brine-to-fish ratio, brining time, brining temperature, fillet thickness, species of fish and quality character (texture or fat content), smoker/drier loading equipment/procedures, hot smoking/drying time and temperatures, humidity control, and character of the smoke and/or hot air circulation in the unit (i.e., *temperature distribution*). There may be other variables depending on your product and process.

Establishing reasonable, conservative test conditions is an important consideration. Doing so may minimize potential critical factors that affect achieving the proper process, may reduce the monitoring effort, and may reduce deviations. Whatever test conditions are settled on, it is critical that those conditions can be consistently applied to actual production.

In all phases of the process validation study, one must carefully document *all* process test conditions, as they will serve as the basis for establishing appropriate prerequisite processing standard operating procedures and/or HACCP controls (critical limits, monitoring, etc.) for the curing and drying/hot smoking process.

The process parameters that should be included in the study design phase, and hence monitored and documented during the study, are those parameters associated with the two major process steps in the production of hot-smoked products that impact achieving the two critical finished product attributes necessary for controlling non-proteolytic *C. botulinum* (i.e., minimum percent WPS and minimum internal fish time/temperature is 30 minutes/145°F). These parameters are summarized in the table below.

Regarding the hot smoking/drying step, it is important to characterize ahead of time the temperature distribution (TD) within hot smoking/drying unit. The reason for this is to identify any cold spots in the unit that may impact achieving the minimum internal product temperature needed for the given process time. The hot smoking/drying process also greatly influences the final moisture of the product, which

Wet curing/brine step	Hot smoking/drying step
Min. strength and/or concentration.	Min. temperatures at each stage, and in various places within the unit.
Min. brine time.	Min. time at each stage.
Min. brine-to-fish ratio.	Using product loading configuration and racks representing worst-case conditions with respect to product heating.
Coldest brine temperature.	
Thickest fillet unit.	
Same species of consistent quality.	

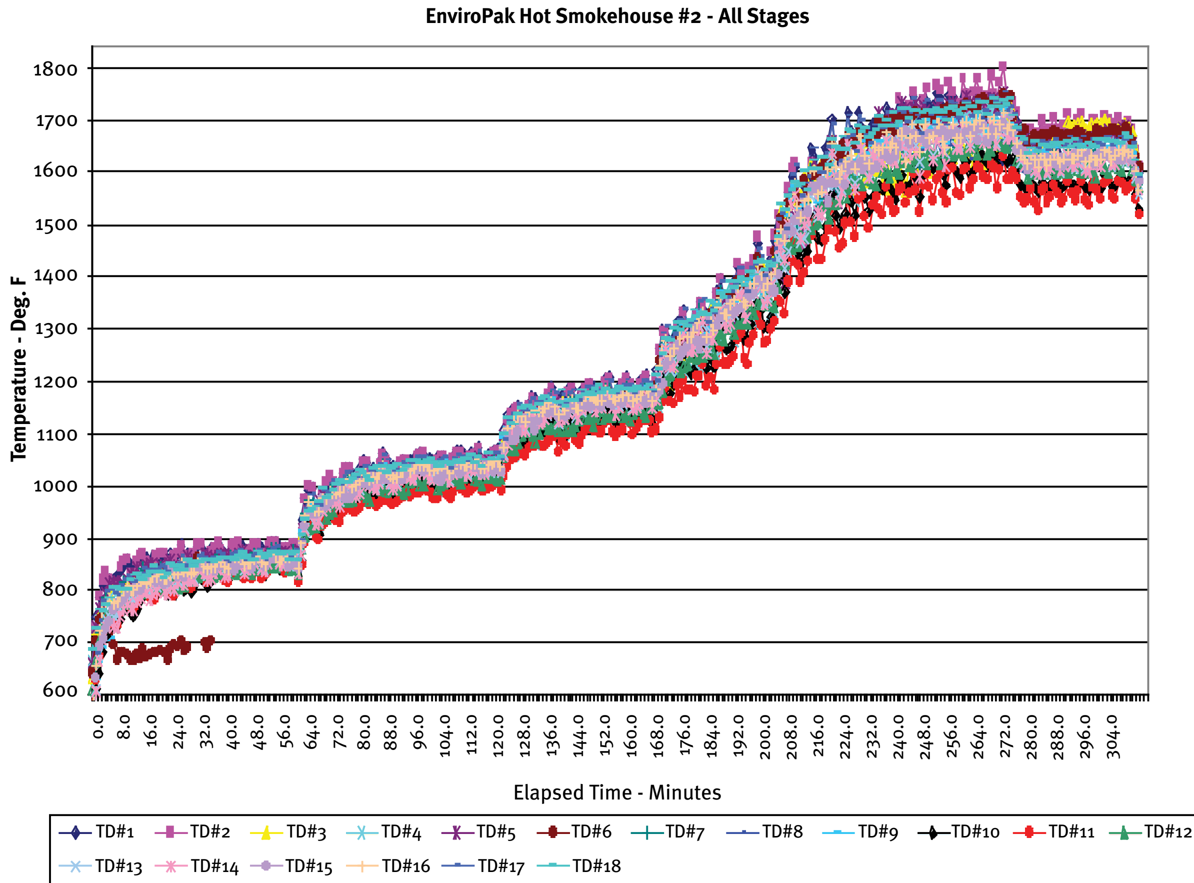

Figure 1. Temperature distribution profile of a hot smoking/drying process (all five stages, start to finish).

directly relates to the ability to achieve proper percent WPS (or a_W). It is important that TD testing of the unit be done *prior to study implementation*, and under full-load conditions, since the data collected with this effort will tell you where the internal temperature monitoring probes (or thermocouples, TCs) should be placed when conducting the actual process validation study. In this regard, the internal temperature monitoring TCs should be placed into product that is (a) representative of the slowest heating fillet piece (generally the thickest piece), and (b) located in the cold region of the unit.

Fig. 1 shows an example of the temperature distribution profile for an actual hot smokehouse. In this particular process, there are five distinct stages of heating and smoke application. Fig. 2 shows only the temperature distribution profile for the latter stage 4 of the process. The data depicted in this graph show that there are a couple of TCs (numbers 10 and 11) with consistently lower temperatures within the unit than the others. For this test, the location of these low reading TCs happens to be in the middle to upper fish racks located in the center of the smokehouse unit. Fig. 3 shows only the temperature distribution profile of the final stage of hot smoking/drying. During this stage of the process, the primary objective is to raise the internal temperature up to the minimum 145°F for 30 minutes. Similar to the data displayed in Fig. 2, these data now clearly demonstrate that the same low-reading TCs (numbers 10 and 11) depict the location of the coldest heating region within the smokehouse. Therefore, it is in these areas of the smokehouse that the temperature probes should be place when conducting the process validation study.

The TD data depicted in all these figures are fairly typical of the temperature ranges experienced in such systems. Processors should work with the equipment manufacturer to ensure their hot smokehouse systems are adjusted and maintained to establish optimal operating conditions and circulation.

Now that the study design has been completed, including the preliminary TD testing, step 2—conducting the validation study—can be completed. This involves gathering process and product attribute data, under the *specific set* of process test conditions identified in your study design phase. Different, separate studies may be necessary depending on your various process conditions, or changes to those

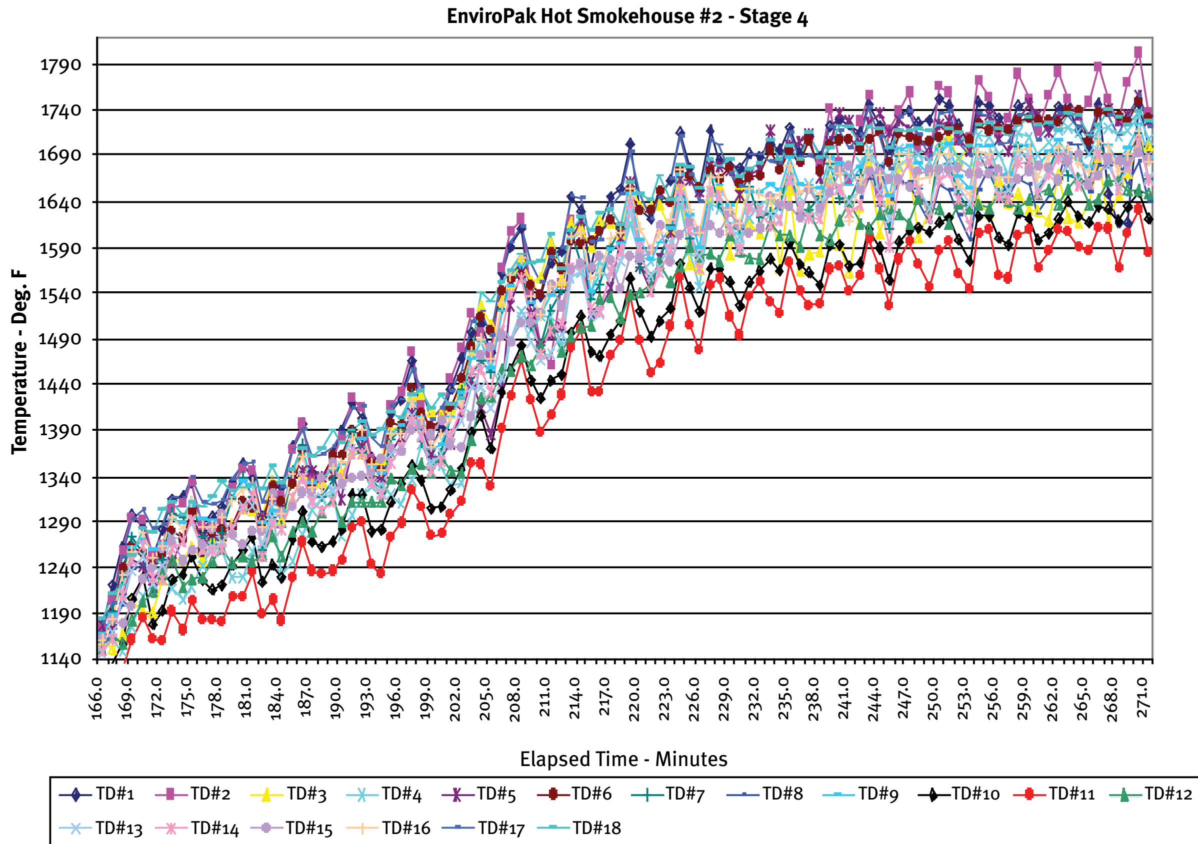

Figure 2. Temperature distribution profile of the latter stage 4 of a hot smoking/drying process.

conditions identified in the study design phase. During the study, it is important that you can track the fillet test units through the entire process, and that the fillet test pieces are placed where they need to be in the hot smoking/drying unit; i.e., the coldest heating region within the smokehouse as determined by the preliminary TD test. It is also critical to keep complete records of all process conditions, in-process and finished product attribute testing (percent WPS, nitrite levels, a_w, etc.), as this information will be considered later on when establishing the necessary HACCP controls and parameters for the process.

Once the validation study has been completed, the data must be analyzed (step 3). Review and analyze the data gathered from the study and the associated study records that were kept. Do they demonstrate that the process achieved the necessary final product attributes, to adequately control non-proteolytic *C. botulinum*? Were the proper percent WPS and/or nitrite levels achieved? Was the proper (minimum) internal core fish temperature reached and was it held there for the proper length of time? If so, then you are ready to move to the final step. If not, then the study design should be re-evaluated, changed accordingly, and implemented again, until it demonstrates that the process can consistently achieve the necessary final product attributes to adequately control non-proteolytic *C. botulinum.*

For the final step, you must use the information from the actual study test conditions to determine the HACCP controls—critical limits, monitoring procedures, etc.—to ensure the process consistently delivers the final product attributes necessary for control of non-proteolytic *C. botulinum*. FDA Guidance (Chapter 13) (FDA 2001) recommends two critical control points for hot-smoked fishery products:

1. Curing step: establish critical limits for, and monitor, the "critical aspects of the established brining, dry salting, and drying process; and the hot smoking process," *or* "the % WPS and/or nitrite levels, where appropriate"; and
2. Drying/hot smoking step: establish critical limits for, and monitor in the final product—"the internal product temperature at the thickest portion of three of the largest fish in the smoking chamber."

The example in the FDA Guide (FDA 2001) recommends that critical aspects of the curing step be monitored for each batch, and the internal product temperature for each batch. Verification of the process would be accomplished by fin-

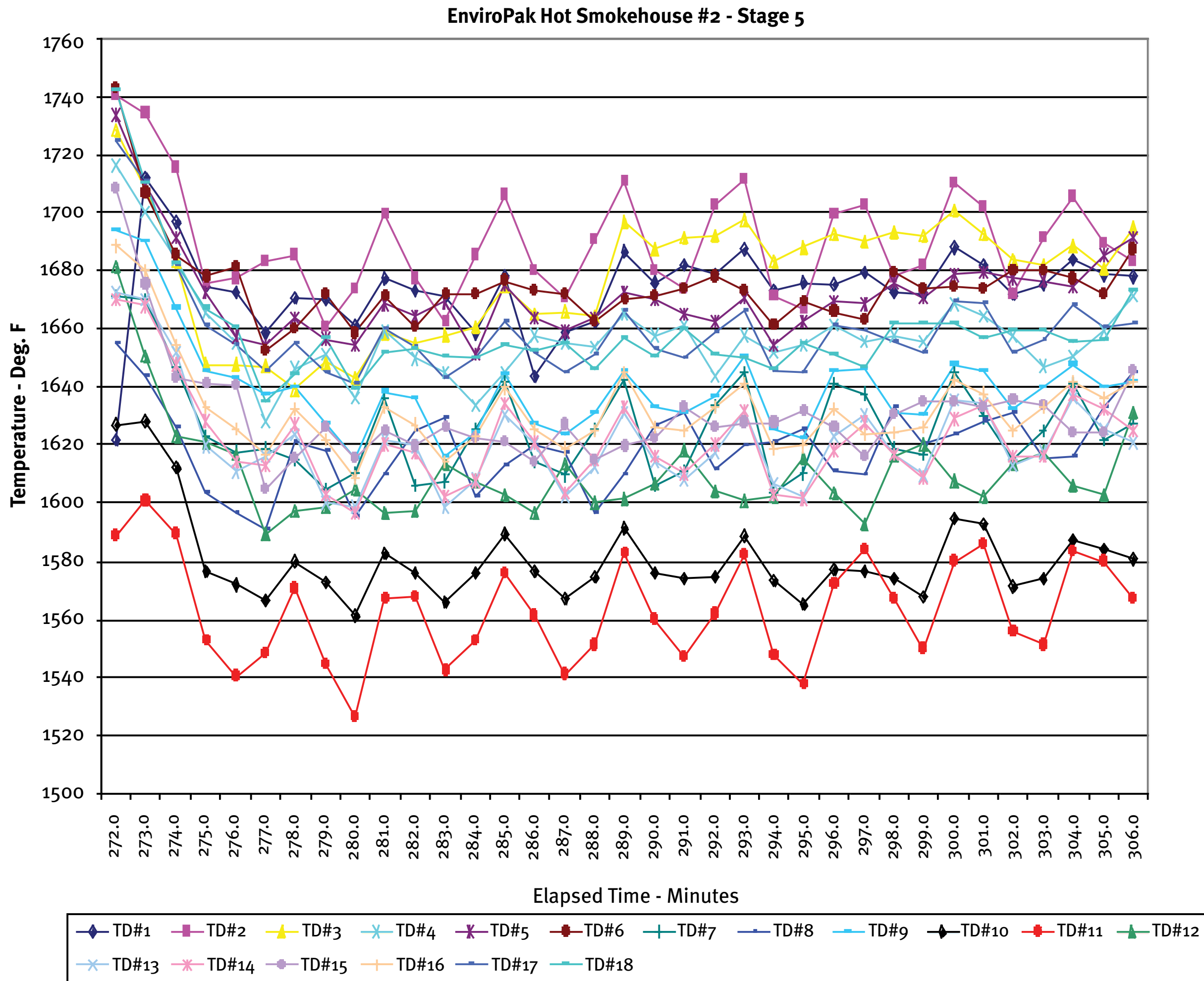

Figure 3. Temperature distribution profile during the final stage 5 of a hot smoking/drying process.

ished product testing to determine percent WPS at least every three months (except where such testing is performed as part of monitoring). Although not mentioned in the FDA Guide, as previously mentioned, FPA recommends that the a_W of the finished product be monitored as well.

If your process establishment/validation study has been designed and performed properly, *and* you have set up appropriate prerequisite processing procedures (for curing, drying, smoking, etc.), then monitoring the percent WPS and internal product temperature of each batch constitutes basically a method of continuous process verification. Other HACCP considerations would include:

- Making sure your prerequisite production operating procedures describe in detail those procedures that will ensure the critical limits and finished product attributes are met.
- Monitoring your process and product often enough to detect normal variability in the values being measured; this is especially true if measured values are close to the critical limit (e.g., percent WPS or a_W).
- Documenting the reasoning used to determine monitoring frequency, number of product units measured, etc.
- Documenting and evaluating changes to the curing and drying/smoking process, to determine the impact of such changes on meeting the critical limits and finished product attributes. Examples are changes to the product preparation and curing step, changes to the drying/smokehouse equipment or loading systems, etc.

Processors who conduct a routine review of the HACCP plan will help identify the need to do additional validation studies for any process changes made.

Although this presentation has focused on hot smoked fishery products, for a refrigerated, vacuum-packaged (reduced oxygen packaged) *cold*-smoked fish product, the

same principles and protocol will apply when designing a study to establish or validate a process, as follows:

- Cure step considerations
- Drying and/or cold smoking considerations
- Ensuring the process achieves the following final product attributes:
  - A 3.5% WPS in the fish flesh, *or* 3.0% WPS + 100 ppm nitrite in the fish flesh, *and*
  - Dried/smoked at a temperature of <90°F.

In the end, for this style of product, it is the interplay of smoke, salt, temperature (low temperature in this case, <90°F), nitrites (if used), *and the presence of competitive spoilage bacteria*, that act as barriers to non-proteolytic *C. botulinum* growth.

GMA/FPA has published additional guidance on this topic, *Establishing or Verifying a Heat Process for Cooked, Ready-to-Eat Seafood Products, and Heat Process Monitoring Considerations Under HACCP* (GMA/FPA 2005). This publication can be ordered at http://www.fpa-food.org/shop/publications.asp.

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Common Mistakes in HACCP for Hot Smoked Salmon

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The U.S. Food and Drug Administration (FDA) contracts with the Alaska Department of Environmental Conservation (ADEC) to perform HACCP inspections throughout Alaska, especially in difficult, remote, or excessively expensive destinations. Dutch Harbor is on Unalaska Island at the top of the Aleutian Chain in the Bering Sea and with the weather, distance, recent discontinuation of jet service to the island, and short runway, this location qualifies as a difficult destination. In the past four years about 30% of the FDA contracts have been in Dutch Harbor.

Year	FDA contracts	Dutch Harbor inspections
2003	283	95
2004	361	98
2005	307	92
2006	353	123

Most of the Dutch Harbor inspections are on vessels that visit the port on sporadic schedules based on fishing openings and offloading schedules, adding another layer of inconvenience to arranging FDA inspections by FDA personnel. ADEC has an environmental health officer stationed in Dutch Harbor.

Anecdotal information was collected during inspections in Dutch Harbor between October 2000 and June 2003, regarding the most common mistakes in formulation and implementation of Hazard Analysis and Critical Control Point (HACCP) plans. This information was published by Alaska Sea Grant in a series called "Common Mistakes in HACCP" in 2004, 2005, and 2006. Each publication is intended to supplement HACCP training and is set up as a list of mistakes and the appropriate corrections. Some issues are specific to Alaska regulations and those outside the state should verify with their own state health agency. Three titles are relevant to smoking seafood.

Hot Smoked Salmon

1. **Not correctly calibrating the thermometer.**
   The recording cook probe thermometer must continually monitor smoking oven temperatures and must be visually verified at least once each batch. Calibration should use a National Institute of Standards and Technology (NIST) traceable thermometer in an ice water slurry for freezing point, and boiling water for boiling point. All calibrations must be documented.
2. **Incorrect placement of the thermometer.**
   The thermometer should be placed in the coldest spot of the smoking oven, which will be determined by a thermocouple study, and then in the thickest piece of fish.
3. **Not having a heat distribution study.**
   Each smoking oven should be checked by a process authority, manufacturer, or independent study to determine cold spots before use and after any modification.
4. **Listing the wrong hazard in frozen product.**
   Pathogens should be controlled through good Sanitation Standard Operating Procedures (SSOPs) and the heating step. Therefore, they are not reasonably likely to occur, which is part of the definition of a hazard.
5. **Not evaluating all the hazards in a refrigerated product.**
   A complete hazard analysis for refrigerated vacuum-packaged smoked salmon needs to evaluate parasites, pathogen growth, *Clostridium botulinum* growth and toxin production, metal inclusion, and allergens/additives.
6. **Incorrect labeling of refrigerated products.**
   Label must specify, "Keep refrigerated below 38°F."
7. **Allowing cross-contamination potential.**
   The flow of product throughout the plant must

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separate finished from raw products to avoid contamination with pathogens, especially after the heat step where the competing spoilage bacteria has been eliminated.

8. **Inadequate water phase salt (WPS) testing.**
The tests must be on a schedule and well-documented. If WPS is not being controlled, the product must be frozen and labeled appropriately.
9. **Incorrect WPS levels.**
10. **Misusing time temperature indicators (TTI).**
If TTIs are used, instructions must be included on each package.

Sanitation Standard Operating Procedures

1. **No written SSOP.**
The federal regulation says you should have a written sanitation plan. The Alaska regulation says you shall have a plan. Neither one says you shall have a written plan but it's pretty clear that you should.
2. **Inadequate record-keeping.**
Both the federal and Alaska regulations require sanitation monitoring records.
3. **Not reading the regulation.**
4. **Omitting any of the eight points of sanitation.**
5. **Ignoring additional water safety aspects.**
In addition to a valid disinfection program checks should be made periodically and documented for back-flow prevention devices and cross connections. Ice production, storage, and delivery should be included in the water safety sanitation point.
6. **Not following the SSOP.**
7. **Not updating in a timely fashion.**
The SSOP needs to be current in both the description of the cleaning procedures and the chemicals used.

Government agencies

1. **Thinking a HACCP plan is required.**
A hazard analysis is required. Only when the hazard analysis reveals hazards that are likely to occur is a HACCP plan required.
2. **Requiring a training certificate.**
For the FDA, the plant HACCP expert may be self trained so long as the knowledge can be demonstrated to the inspector. However, if the plant is participating in the USDC HACCP/Quality Management Program personnel must successfully complete the USDC HACCP training.
3. **Review by an unqualified person.**
A HACCP-knowledgeable person must review the records within one week.
4. **Not reading the six pages of HACCP regulation, 21CFR parts 123.6-12.**
5. **Not reading the Good Manufacturing Practices, 21CFR part 100.**
6. **Confusing the agencies who deal with HACCP.**
FDA regulates the production and distribution of seafood and their HACCP program is about seafood safety only.

   USDA (U.S. Department of Agriculture) inspects meat from land animals and have a HACCP program for meat.

   USDC (U.S. Department of Commerce) offers a fee-for-service seafood inspection program that includes economic integrity, quality, and wholesomeness along with seafood safety in their seafood HACCP program.

   EPA (Environmental Protection Agency). The only connection with HACCP is that EPA sets tolerance levels for certain food contaminants.

   ADEC. Along with the health inspection agencies in other states, ADEC has adopted the federal seafood HACCP rule as well as imposing several rules unique to Alaska.

These mistakes were commonly made in HACCP plans on vessels in Dutch Harbor, and may help processors who will soon be writing or updating their own plans. Knowing these mistakes may help those who are performing hazard analyses and formulating HACCP plans, but processors should be aware that regulations change and inspectors' interpretations may sometimes vary.

For more details and for other titles in the "Common Mistakes in HACCP" series, go to the Alaska Sea Grant bookstore at http://seagrant.uaf.edu/bookstore/pubs/ASG-38to41.html.

Wood Smoke Components and Functional Properties

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Abstract

Wood smoke is an aerosol produced by pyrolysis of wood at elevated temperatures and reduced oxygen. Generated wood smoke consists of three phases: compounds in the gaseous phase, particles of liquid droplets, and solids. When condensed and given enough time for the polymerized components to settle out, the liquid fraction is called liquid smoke. There are over 400 compounds identified in wood smoke or smoke flavor from a number of sources. So far, 40 acids, 22 alcohols, 131 carbonyls, 22 esters, 46 furans, 16 lactones, and 75 phenols have been identified. The origin of the compounds in wood smoke is the polymers in the wood and the heat-induced chemical reaction between the heated polymers, gasified intermediates, and moisture. Thus, the composition of wood smoke will vary with the type of wood used in producing the smoke and the temperature and moisture content of the wood.

Wood smoke performs several functional roles in food. Whether it is applied as a gas from smoldering wood chunks or chips or as liquid smoke, it is considered a natural flavor and need not be broken down into components in the label declaration. Wood smoke is also a colorant, where the stain is immediately produced upon contact between the food surface and smoke, or the color is formed when the smoke and food components react chemically at the elevated temperature used to process the food. The preservative role of wood smoke is well known. However, although specific components have been documented to possess inhibitory activity against bacteria and fungi, wood smoke is not a stand-alone preservative. Wood smoke may be used as a component of a hurdle system for food preservation. Among the functional components of smoke, phenols and acids have shown the most antimicrobial activity, although there are data that show that carbonyls and acids can also have a wide spectrum of antibacterial activity even at low levels of phenols. Staining ability of wood smoke is associated with the acids and phenols, while reaction-developed color produced during the heating of meat can be attributed primarily to the acids and carbonyl compounds.

Introduction

Direct exposure of fish to smoke generated by a smoldering wood fire is a process that has been used since ancient times. Before the arrival of Europeans in the New World, Native Americans had been smoking fish to preserve them for consumption on long treks or later in the season when these fishes were no longer available in the wild. For true preservative effect, smoke has been combined with water activity reduction by dehydration and salting. More recent is the use of smoke for flavor. Traditional cured meats used to be cooked in smokehouses where dehydration occurs in addition to smoke deposition. As smokehouses have become more modern, the processes of smoke flavor application and dehydration can be separated with better control of both by the processor. There have been claims about the desirability of smoke generated from certain species of wood compared to other woods, but there have been no studies of side-by-side comparison of flavor imparted by a smoke from specific woods on a smoked product.

In general, smoke is generated using wood that is readily available in a locality. Thus, smoke is generated from mesquite in the Southwest, hickory, oak, and wild cherry in the South, apple wood and maple in the Northeast, and alder, birch, and beech in the Rocky Mountain region and the Northwest. A recent trend in the smoke flavor industry is to produce liquid smoke as a byproduct of the process of making briquetted charcoal for backyard barbecues. Condensed smoke or smoke dissolved in water may be obtained by generating smoke using one type of wood or a mixture of different woods, then the liquid smoke composition is standardized depending upon the application. This review covers compounds present in smoke and the desirable smoke components for specified functional effects in the finished product.

Physical nature of wood smoke

Wood smoke produced by heating wood chunks, chips, or sawdust is a colloidal aerosol of air, water vapor, solid particles, liquid droplets, and vaporized organic compounds. The vapor phase of the organic compounds in smoke imparts the desirable flavor since rate of deposition in the food is slow and uniform. Preferably, the liquid phase is not directly deposited. This phase serves as a reservoir for generation of more of the vapor phase as the smoke temperature increases. The best smoke generators route the smoke under a curtain of flowing water to remove solid phase and liquid phase particles before

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the smoke enters the smokehouse. Solid particles are associated with soot, tar, and fly ash and are best eliminated before the smoke enters the smoke house. Tilgner (1976) suggested that liquid smoke components can also polymerize to form solid particles. Thus, depending upon the smoke temperature entering the smokehouse and the temperature of the smokehouse itself, there could be tar formation from smoke even if most of the tar has been removed prior to the smoke entering the smokehouse. The visible component of smoke consists of liquids and solids in the aerosol.

Foster and Simpson (1961) showed that the solid and liquid component of smoke do not contribute to the desirable smoke flavor. However, in the absence of analytical instruments that could measure the vapor phase organic compounds in smoke, one way to ensure that adequate smoke components contact the product in the smokehouse is to ensure that a dense optically visible smoke is generated by the smoke generator. An alternative is to generate gaseous smoke from liquid smoke. This can be done by atomizing liquid smoke into the warm smokehouse air as the air is recirculated, or by dripping liquid smoke on a hot plate located just at the point where make-up air enters the smokehouse. These techniques ensure that there are adequate organic compounds from smoke in the smokehouse atmosphere that could be deposited on the product. Recent environmental regulations related to the release of volatile organic compounds (VOCs) into the atmosphere have curtailed the use of smoke generators in processing plants located in metropolitan areas. Smokehouses in these areas have to be equipped with an incinerator to eliminate VOCs in gas discharged from the smokehouse.

Wood smoke composition

Most of the literature on wood smoke composition is based on the components of smoke condensate generated in a controlled laboratory setting. Wasserman and Fiddler (1969) reported approximately 20 times the level of compounds in the condensate compared to that in the gaseous smoke. A number of compounds with long elution times are present in the smoke condensate but were absent in chromatograms of gaseous smoke, indicating that some polymerization of smoke components may have occurred in the liquid phase. These same authors also showed that smoke condensate improved in flavor with aging such that the 5 hour old condensate has the worst flavor while the 1 and 2 months old smoke had similar and the highest acceptability scores. The classes of compounds and specific compounds identified in wood smoke reported by Wasserman and Fiddler (1969) are shown in Table 1. These same authors also report that the concentration of the furans, phenolics, and cyclic compounds are affected by the excess oxygen supplied to the burning wood, and that formation of phenols and cyclic compounds

Table 1. Classes and examples of compounds predominant in wood smoke.

Class of compound	Examples
Acids	Acetic acid
Alcohols	Methanol, ethanol, isopropanol, furfuryl alcohol
Carbonyls (aldehydes)	Acetaldehyde, isovaleraldehyde, crotonaldehyde, diacetyl
Carbonyls (ketones)	Acetone, methyl vinyl ketone, acetol, 2-cyclopentnone, furfural, 5-methyl furfural, cyclotene
Esters	Methyl formate, methyl acetate
Furans	Furan, 2-methyl furan
Phenols	Phenol, 1,2-dimethoxybenzene, guiacol, 4-methyl guiacol, 4-ethyl guiacol, eugenol, syringol, 4-methyl syringol, 4-ethyl syringol, 4-propyl syringol, vanillin

Source: Wasserman and Fiddler 1969.

Table 2. Composition of different woods and the chemical constituents of condensed smoke.

	Wood composition (%)		Smoke composition (%)			
Wood	Lignin	Cellulose and hemicellulose	Phenolic and basic	Acid	Smoke pH	Smoke point °C
Cherry	13.8	24.1	46	18	2.46	147
Red oak	24.1	62	62	27	2.53	135
White oak	39.3	25	61	22	2.67	122
Hickory	24.1	43.1	55	27	2.71	103
Apple	37.9	27.6	47	22	2.72	103
Hard maple	55.2	34.4	45	18	2.74	107
Chestnut	32.1	25	34	44	2.84	121
Mesquite	44	16	44	12	3.02	164

Source: Chen and Maga 1993.

Table 3. Groups of compounds and percentage of chromatograph peak area in condensed smoke from a charcoal kiln.

Class of compound	Most predominant	% of peak area
Acid	Acetic acid	13.9
Alcohol	Methyl alcohol, acetol	13.5
Unidentified	–	12.7
Phenol	Guiacol, 2,6,dimethoxy phenol	9.3
Ketone	1-hydroxy, 2 butanone	5.2
Hydrocarbon	Cyclotene	3.1
Aldehyde	4-hydroxy-3,5 dimethoxy benzaldehyde	0.8

Calculated from product analytical data, Hickory Specialties Technical Center, Brentwood, Tenn.

Table 4. Composition of wood smoke from 14 and 37% moisture alder wood.

	Composition (%)	
Compound	37% H_2O	14% H_2O
Acetic acid	49.5	94.5
Propionic acid	10.1	1.9
Butyric acid	3.7	0.2
Valeric acid	2.8	0.2
Caproic acid	3.7	0.2
Hydroxypropanone	3.1	0.5
1-acetoxy-2-butanone	0.8	0.04
2-furfural	13.7	1.24
5-methyl-2-furfural	0.7	0.07
2-methoxy-4-methylphenol	2.5	0.09
2,6-dimethoxyphenol	1.5	0
2-methoxy-4-transpropenylphenol	1.7	0.07
2,6-dimethoxy-4-methylphenol	3.1	0.6
2,6,dimethoxy-4-allylphenol	1.6	0.2
Vanillin	1.1	0.03
Benzo(k)fluorantene	0.02	0.007
Benzo(a)pyrene	0.008	0.0003
Pyrene	0.4	0.1
Chrysene	0.05	0.03

Calculated from regression equations reported by Borys et al. 1977.

is favored at around 10 to 12% excess oxygen while formation of furans is favored at near zero excess oxygen.

Composition of a commercial liquid smoke preparation produced by water absorption of gaseous smoke from a charcoal kiln is shown in Table 2 (Hickory Specialties, Brentwood, Tennessee, pers. comm.). There were 61 gas chromatogram peaks reported in the sample. Twenty peaks were not identified. The percent of peak area indicate that acids, alcohols, and phenols predominate in the condensed smoke. Compared to phenol (mol. wt. 94) retention time of 26.4 min, the unidentified peaks were mostly compounds with retention times less than 20 min with molecular weights estimated to be in the 90 to 160 Dalton range.

Since the origin of chemical components of wood smoke is in the wood itself, the type of wood and the method of generation would be expected to affect smoke composition. Chen and Maga (1993) compared properties of wood smoke produced from apple, cherry, chestnut, hard maple, hickory mesquite, red oak, and white oak. Table 3 summarizes their data. No direct correlations can be made on polymer fraction in the wood and the composition of smoke generated. The wood shavings were not adjusted for moisture content so moisture is not a factor in these analyses. The smokepoint of the wood increased with decreasing concentration of combined cellulose and hemicellulose, and was higher as the lignin content increased. Concentration of acidic components in the smoke condensate did not correlate with pH of the liquid smoke. The most desirable smoke flavors are derived from phenolic compounds and compounds with a basic reaction (Maga 1987), but the concentrations of these compounds did not correlate with the lignin content of the wood. The highest concentration of phenolic and basic components in the smoke condensate were produced by red oak, white oak, and hickory while the least were produced by mesquite and chestnut.

The influence of moisture content of alder wood used to generate smoke was reported by Borys et al. (1977). Their results were presented as a regression equation of the gas chromatogram peak areas of specific compounds, as a function of the rate of aeration of the wood during smoke generation. Table 4 shows the composition of wood smoke from wood at 14% and 37% moisture. At the low moisture content the smoke is primarily acetic acid, which constitutes 94% of the smoke. The more desirable smoke was generated from 37% moisture wood where the compounds are more widely distributed. At this moisture content the acids account for 70% of the smoke while carbonyls and phenols account for 18 and 11.5%, respectively. On the other hand, smoke generated from 14% moisture wood had only 2 and 1% respectively of carbonyls and phenols. The results of this work justify the widely used practice of wetting sawdust or wood chips used in smoke generators.

Kjallstrand and Petersson (2001) found 2,6-dinitrophenols as the compound with the highest concentration in alder smoke. 2-methoxy phenol, 2,6-anhydroglucose (furfural), 2-furaldehyde, 2-methylfuran, benzene, and methylbenzene were the other compounds with significant concentrations in the smoke. Acids, alcohols, and ketones were not reported.

A lot of variability in smoke composition is reported in the literature. Some of the sources of variation are the type and particle size of wood used, the moisture content, the rate of aeration during smoke generation, and the temperature of the wood during pyrolysis. Maga (1987) conducted a thorough review of the flavor chemistry of wood smoke and a

Table 5. Compounds in wood smoke.

Compound	Number identified	Examples
Acids	48	Acetic, propionic, sorbic, butyric, salicylic, benzoic, pimaric
Alcohols	22	Methanol, ethanol, isopropyl, cyclohexanol, benzylalcohol
Carbonyls	131	Ethanol, acetone, methylethylketone, acetoxypropanone, 2-heptanone
Esters	22	Methylacetate, ethylbenzoate, hydroxy-2-propanone propionate
Furans	46	Furfural, benzofuran, 2-methyl-3-furfural, 2-acetyl-5-methylfuran
Lactones	16	γ-butyrolactone, methylvinyl-2-butenolide
Phenols	75	Phenol, xylenol, syringol, eugenol, resorcinol, 4-isopropylguiacol
Miscellaneous	50	Pyrazine, pyridine, toluene, ethanediol, pyrrole, dimethoxyethane

A comprehensive list of the compounds is given in Maga 1987.

brief summary of his compilation of compounds whose presence has been identified in wood smoke shown in Table 5. Not all published data on smoke composition reported by different authors have the same compounds listed. Furthermore, the concentration of the different compounds varied among these studies. Since flavor is a complex interaction between components, actual perceived flavor could vary even with the same components present if the relative concentration of the different components is different in the different smoke samples. Thus, it is important that the method for smoke generation be standardized. Consistency of smoke composition is assured by using liquid smoke where the manufacturer has standardized the product composition.

Ensuring consistency of smoke application from gaseous wood smoke

Gaseous smoke composition can only be consistent if the same smoke generator is used and the same conditions are used in the smoke generator. Modern smoking ovens use a separate unit for smoke generation and smoke application to the smoked product. This is in contrast to simple ovens where the smoke is generated from smoldering wood placed directly under the racks that hold the product. In these simple smoking ovens, it will be difficult to control smoke generation temperature, oven temperature, and humidity. When using smoke generators, uniform smoke intensity and smoke composition can be obtained by ensuring that (1) the type and particle size of wood is the same, (2) maintaining a constant moisture content by mixing a batch of wood chips adequate for the time required for the smoking process with water to the desired moisture content, (3) the orifices that feed air to the wood in the generator are clear, (4) the wood is burning uniformly around the periphery of the burning zone before moist sawdust is added, and (5) the wood is added at rates that will maintain the same thickness of the unburned wood over the burning layer. Adding wood chips too fast will result in a thick layer of moist unburned wood over the fire zone, stifling combustion and eventually extinguishing the fire zone. On the other hand, adding the wood chips slowly will expose the fire zone, generate heat, and make the wood flame up, reducing the intensity of smoke generation. These procedures are consistent with the factors discussed above on the factors that affect the composition of wood smoke.

Smoke flavors

The term liquid smoke generally refers to the condensate of wood smoke. Smoke from a charcoal kiln is directed through a flue into an absorption tower where a liquid water film flowing countercurrent to the flow of smoke captures the smoke. The smoke solution is held for several days to permit the condensed phenolic compounds to precipitate and the solution is filtered. The process removes polymeric aromatic hydrocarbons, which are carcinogenic. Thus, the use of liquid smoke imparts both convenience and safety.

A number of wood smoke preparations are now used in the industry as a flavor. Smoke flavor is GRAS (generally regarded as safe) and is considered by both the USDA and FDA as a natural flavor. Liquid smoke can be applied as a dip or a drench to color and flavor cooked ready-to-eat meats, and added directly to other food products such as barbecue sauce, dry crispy snacks, canned baked beans, and canned fish and shellfish. In metropolitan areas where discharge of smoke to the environment is restricted, liquid smoke may be used to generate gaseous smoke.

Knowledge of the composition of smoke has helped the industry to produce smoke preparations with different flavor and functional properties. Since flavor is a result of the interactions among the various compounds in smoke, treatments that remove certain smoke components may be used to alter the flavor of smoke. Thus, vacuum evaporation may be used to remove low boiling components such as acetic acid and alcohols. Adsorbents may be used to selectively remove phenolic compounds and more importantly, remove condensed phenolics that produce a tarry precipitate in stored liquid smoke. The solubility of smoke in water can be enhanced by adding polyethylene glycol to the liquid smoke. The presence of polyethylene glycol also minimizes condensation of phenolics. Acids may be neutralized to reduce the harshness of the smoke flavor. Dekker (2003) interviewed a major liquid smoke manufacturer in the United States and discussed how liquid smoke flavors are produced and how the composition is standardized. In addition to the standard smoke condensate, products may be made from fractions of the liquid smoke produced by selectively extracting components with an appropriate solvent. For example, an oil extract of liquid smoke will contain primarily phenolics with the desirable smoke flavor, while the harsh flavored acids and carbonyls

are not transferred to the oil. The smoke flavored oil will also induce minimal color development in the product during heating because reduced levels of carbonyl compounds have been transferred to the oil from the liquid smoke.

Some applications of smoke flavor may require a solid phase material. An example of this is smoke flavored salt used on oven-baked cooked meat to simulate a grilled or broiled flavor. To produce the solid smoked flavor, liquid smoke is plated on a soluble solid carrier such as salt or maltodextrin.

Functional properties of smoke components

Flavor

It is generally recognized that the characteristic wood smoke flavor is due to phenolic compounds (Bratzler et al. 1969, Deng et al. 1974, Maga 1987). Thus, the concentration of phenols has been used to assess the intensity of the smoke flavor (Chan et al. 1975, Kjallstrand and Petersson 2001) in smoked fish or meats. However, flavor is a complex sensation that is imparted by combinations of different compounds. Thus, a single group of compounds may not necessarily completely bring out the smoke flavor. The contribution of the acids and carbonyls to flavor is important, although these compounds may be present in smaller amounts than the phenols. When liquid smoke is separated into the phenolic, carbonyl, and non-carbonyl fractions and a new liquid smoke mixture is made, the aroma quality of the mixture is best when the phenolic fraction is mixed with the appropriate amount of carbonyl and non-carbonyl fraction (Maga 1987).

Some of the phenols in smoke are similar to those in spices. An example is eugenol in cinnamon, pepper, nutmeg, marjoram, and cloves. Cinnamon contains many of the phenolic compounds present in smoke. The flavor of smoke components also depends upon the concentration. Thus, the same smoke flavor may invoke sensory responses of burnt, pungent, and cresolic at high concentration or sweet smoky at the desirable concentration. These data in the literature suggests that not all liquid smoke preparations are the same in terms of flavor and other functional properties, and that each application will benefit from a careful selection of the right smoke flavor.

Color

Color formation in smoked products is due to a combination of cold staining and heat-induced Maillard-type chemical reactions. When using liquid smoke, a cold stain is imparted by the phenolics and acids. When the product is heated, the carbonyl compounds react with the proteins in a Maillard reaction to produce the brown color. Cold smoking is the term used when raw fish is exposed to a relatively low optical density smoke at temperatures below the denaturation temperature of fish muscle (below 40°C). Cold smoked fish does not change in color because there are not enough phenols to produce a stain and the Maillard reaction does not proceed far enough to develop the color. On the other hand, smoking at temperatures of 80 to 90°C results in excessive deposition of phenolics, formation of condensed phenolics, and excessive Maillard browning so that the product has a dull dark color with numerous small specks of dark material on the surface. The best smoked fish color is a glistening golden honey color brought about by the capture of phenolics on an oily surface and adequate Maillard reaction to generate a light brown color (Deng. et al. 1974, Chan et al. 1975). Since smoke can be labeled as a natural flavor, it is a preferred ingredient and one of the applications is a roast color accelerator with no smoke flavor at all. The smoke generated from starch or very low lignin wood is practically devoid of phenolics but is very high in carbonyls; therefore this product can be used as a browning agent.

Table 6. Smoke fractions tested for antimicrobial activity and their properties.[a]

	Smoke extract (%)	Acidity pH	Phenol content (mg/ml)	Carbonyl content (mg/ml)
F1	4.5-5.9	2-3.0	0-5	151-200.9
F2	0-1.4	6.1-7.0	0-5	101-150.9
F3	6.0-7.4	2-3.0	0-5	101-150.9
F4	3.0-4.4	4.1-5.0	20.1-25.0	0-50.9
F5	6.0-7.4	2-3.0	0-5	101-150.9
F6	6.0-7.4	2-3.0	0-5	51-100.9
F7	1.5-2.9	5.1-6.0	0-5	51-100.9
F8	0-1.4	6.1-7.0	0-5	101-150.9
F9	0-1.4	6.1-7.0	0-5	51-100.9

[a]Acidity as acetic acid; phenols as 2,6, dimethoxy phenol; and carbonyls as 2-butanone.
Source: Milly et al. 2005. Analytical data provided by MasterTaste Inc. Zesti Smoke Division, Brentwood, Tenn.

Antimicrobial

The antimicrobial properties of smoke are well known and constitutes the primary role of smoke in food preservation. All the constituents of smoke interact to bring about the antimicrobial effect. Phenolics, acetic acid, and carbonyls individually have antimicrobial activity but their combined effect is synergistic. Thus, a mixture will be an effective antimicrobial agent at a lower level of the components than any of the individual components. Because of the differences in the composition of liquid smoke, inhibitory activity varies with different smoke preparations. Studies on antimicrobial properties of CharSol smoke preparations (Wendorff 1981) at 0.5% was bactericidal to pathogens such as *Escherichia coli*, *Staphylococcus aureus*, and *Listeria monocytogenes*. Some preparations required lower concentration than others for bactericidal activity. It was hypothesized that the enhanced bactericidal activity of some preparations was due to the higher levels of polar phenolic compounds in these preparations (Messina et al. 1988).

The role of carbonyls and acids in the antimicrobial activity of liquid smoke was reported by Milly et al. 2005.

Table 7. Minimum inhibitory concentration (%) of nine smoke fractions shown in Table 5 on different microorganisms.

	Gram negative bacteria[a]	*Listeria innocua* M1	*Lactobacillus plantarum*	*Aspergillus niger*
F1	1.5	1.5	0.75	1.5
F2	>2	2	1.5	2.5
F3	2	>2	2	2.5
F4	3	2	>10	NT
F5	2	2	4	NT
F6	2	2	5	NT
F7	5	4	7	NT
F8	>2	2	>2	>5
F8	9	6	>10	NT

[a]Cocktail of *Salmonella muenster*, *Salmonella senftenberg*, *Salmonella typhimurium* and *Escherichia coli* 8677.
NT = not tested. Source: Milly et al. 2005.

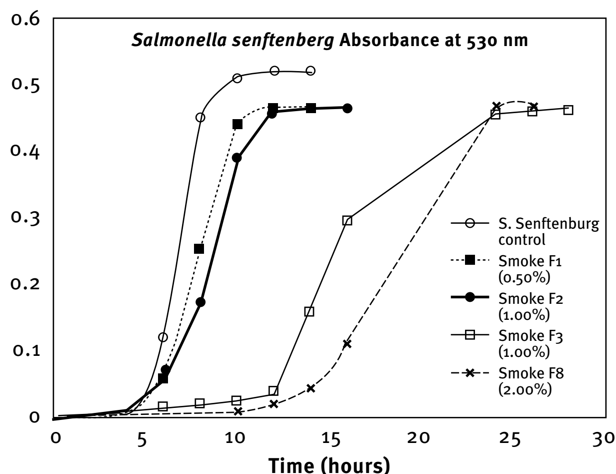

Figure 1. Growth curve of *Salmonella senftenberg* in tryptic soy broth containing liquid smoke fractions F1, F2, F3, and F8 at concentrations below the minimum inhibitory concentration (MIC).

Table 6 shows the composition of the liquid smoke preparations studied by Milly et al. (2005). The smoke preparations vary in acidity from 0 to 7.4% as acetic acid, and carbonyl content from 51 to 200 mg per ml as 2-butanone. Except for one preparation that had as much as 25 mg per ml of phenols as 2,6-dimethyoxyphenol, the rest had 0 to 5 mg per ml of phenols. Minimum inhibitory activity was measured as the smoke concentration in a liquid growth medium that prevented growth of the test microorganism. Table 7 shows the minimum inhibitory concentration (MIC) of the different smoke fractions against several microorganisms. The high phenol, low carbonyl, medium acidity fraction F4 had the highest MIC against *Lactobacillus plantarum*, but had similar MIC as fractions having similar acidity against *Listeria innocua* M1. The fraction with the lowest MIC on all the microorganisms tested was F1, which had medium acidity and low phenol but had the highest carbonyl concentration. The fraction F7, which also required a relatively high MIC, had low acid, phenols, and carbonyl concentration. The smoke fractions also had antifungal activity against *Aspergillus niger* (Table 7). F1 had the lowest and F8 the highest MIC against the mold. The manner in which the smoke fractions affected microbial growth is shown in Fig. 1 for *Salmonella senftenberg* applied to the growth medium at concentrations below the MIC. Below the MIC smoke extended the lag time for growth but once growth started, the organisms increased in numbers at a very rapid rate, not much different from the control. These results show that the carbonyls are a very important component of smoke, not only from the standpoint of color formation but also from the standpoint of antimicrobial activity.

Concluding remarks

Much is known about the composition of smoke and the role of these components in imparting flavor, color, and antimicrobial properties to the smoked product. However, the interaction of the components, the relative concentration of these components, and the level present in the product all affect the flavor. When using gaseous wood smoke, it is important that the conditions used in generating the smoke are maintained constant to ensure consistency of smoke functional properties. When using liquid smoke flavors, it is also necessary to optimize the type and level of smoke flavor for a given application. Antimicrobial liquid smoke preparations containing very low levels of phenols and thus low smoke flavor are now available commercially. These preparations may be used in combination with other antimicrobial treatments to improve the safety of ready-to-eat cooked meats designed to carry very low background smoke flavor.

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Introduction to Smoke Condensates

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Abstract

The smoking of food has been an important tool used to preserve meats for thousands of years. Today it is still utilized to preserve foods and provide a wide variety of flavors to those foods. Historically, traditional smoking has been done with the burning of wood chips at the processor, which provides the smoke but also leads to tar formation in the smoke house, pollution in the environment, and carcinogens on the food. However, there is an option to provide a very good flavor and color, reduce tar and pollution, and make the food safer to eat. This is by using smoke condensates. These are solutions made by burning sawdust in controlled conditions, which allows for flavor selection, tar elimination, pollution control, and carcinogen reduction. The smoke condensates can be applied by various methods and allow for several additional advantages for the food processor.

Smoke condensates

Natural smoke condensates have been produced for almost 50 years. What started as a simple process producing one smoke flavor has turned into a large market with a few manufacturers producing hundreds of different condensates to replace the traditional burning of wood chips. Meat companies in nearly one hundred countries utilize natural smoke condensates to produce high quality, safe, and consistent products. Outside the meat industry, smoke condensates are utilized in numerous types of food items. Manufacturers can create a wide range of smoky tastes, improve overall flavor impact and eating quality of products made under rigorous conditions, and enhance the appearance of ready-to-eat foods.

Natural smoke condensates are produced from the controlled burning of sawdust. The sawdust is collected as a byproduct from the lumber industry. It is sorted by species as it is collected so the smoke generated can be identified by the species from which it was derived. Sawdust generally comes to the smoke manufacturer in semi trailers directly from sawmills. The sawdust has a moisture content of 20-35%. At the smoke condensate factory, the sawdust is carefully dried to reduce the moisture to a consistent 3-4%. It is also screened to obtain a uniform particle size for an even "burn" in the smoke generators. This burning process is called pyrolysis, the controlled chemical decomposition of wood in limited oxygen.

Red Arrow utilizes two different types of reactors for manufacturing smoke condensates, Calciner and Rapid Thermal Processing (RTP). The Calciner method uses indirect heating to warm a rotating drum, which is sealed to prevent air from getting in. Sawdust is introduced into one end of the reactor and free falls in the hot air of the drum as it rotates. As the sawdust particle gets hot, it begins to smolder, sending smoke back up through the drum. The sawdust stays in the drum for about 1 minute as it passes through. When it gets to the end of the drum, it is charcoal. The waste charcoal produced at Red Arrow is used as a fuel source for the sawdust dryer. The smoke vapor that is produced is fed into the bottom of a column where water is recirculated from the bottom to the top and then falls over the rising smoke cloud. All the water-soluble components are absorbed by the water, thus forming a smoke condensate. This material is circulated through the column until a particular concentration is reached, and then is drawn off as new material is formed The raw material is allowed to settle for a few days to allow all the tars and unwanted materials to fall out of solution, leaving behind a clean smoke condensate ready for use. All tar that is collected at Red Arrow is burned as a fuel in the boiler and afterburner to rid the factory of any fumes.

The RTP is a much faster process where superheated sand is recirculated in a reaction column. When the sand is hot enough, sawdust is fed into the stream of superhot sand. This causes an instantaneous pyrolysis of the wood forming the smoke vapor. It then travels on through a column similar to the Calciner to condense the vapor into a smoke condensate. The speed of the two reactors causes different flavors and other organic compounds to be formed.

Traditional wood smoke consists of two parts: the particulate phase, which is visible to the eye, and the gaseous phase. The particulate phase contains fly ash and tars. The tars are the fraction of wood smoke that contain the vast majority of the PAHs, or polycyclic aromatic hydrocarbons. These are the cancer-causing agents. The generation process of smoke condensates greatly reduces, or eliminates, these compounds from getting onto the food product. The gaseous phase consists of acids, carbonyls, and phenolics. These are the components that are desired in a smoke providing the flavor, color, and preservation properties of the smoke. Acid provides the smoke's tartness, contributes to the skin formation of sausages, and accelerates the nitrite cure reaction. Carbonyls are responsible for the coloring capability of smoke. It is a key reactant in the Maillard reaction, which is the chemical reaction that causes foods to brown when heated. From this process, minor flavor components are also formed. The main flavor comes from the phenolics. These ring structures have various side chains that provide dif-

ferent flavors depending on how the smoke was generated. Phenolics are also responsible for the majority of the antimicrobial activity of smoke.

Smoke condensates are generated by two main reactions, but then can be treated in several ways to produce hundreds of different flavor possibilities. Some of these processes are concentration, dilution, resin treatments, different reaction conditions during production, addition of emulsifiers, pH adjustments, blending, and many more. Continuous research allows for improved production of more and better flavors. These flavors can then be found in several forms including aqueous, water soluble, dry powders, and oil based. The product utilized in a food product depends on what one wants to achieve and how the food will be processed.

As with the variety of type and flavor of smoke condensates, there are many ways these products can be utilized in food production. The application method most similar to traditional smoking is atomization. In this process, the smoke condensate is applied using high air pressure, which breaks the smoke condensate into very small droplets. When done correctly, it forms a smoke cloud that is “dry” to the touch and can be circulated in a smokehouse as would traditional smoke. A second application method is showering. In this method, smoke condensate is diluted to a 5-50% solution with water and cascaded over the food product, filtered, and recirculated back through the process. This is most commonly used in hot dog and other large meat production applications. Showering allows for greatly reduced cook times while ensuring uniform color and flavor of the product. Third is direct addition. This is used in almost any food product where a smoky or grilled flavor is wanted. As the name suggests, the smoke condensates are added directly to the meat emulsion or marinade or injected right into the meat. This is used to give a smoky note to foods that are cooked in a bag or in a steam tunnel. Last is topical application. This is used in dry rubs and seasoning blends for foods that are not cooked in the factory or not normally cooked in a smokehouse.

Why use smoke condensates instead of traditional smoking? There are several advantages. The first is flavor and color uniformity. Due to highly controlled specifications of the smoke condensates, the processor is assured that every batch they produce will be identical to the previous batch and that all the products in a smokehouse will be the same. This cannot be said for traditionally smoked items. Since a smoked flavor can be developed without going through a smoke cycle, the production schedule can be greatly reduced. This increases production without adding additional smokehouses. No smoking cycle means less time in the cooker, which means more cook cycles per day, which means more production per day!

The most important advantage of smoke condensate is how clean it is. By not turning on a smokehouse smoke generator, there is no tar formation in the smokehouse. This will reduce cleanup time drastically. No smoke generators also means no need for emission control as there are no, or very limited, emissions. The product produced is also cleaner and safer. Smoke condensate companies remove the tar from smoke, which is the component that contributes the most carcinogenic components to meat. Thus the meat is much safer to eat.

Smoke condensates are regulated by the FDA and USDA. Labeling requirements for the various application techniques are

Atomization	Smoked	(USDA Policy Memo 058A-1)
Drenched and topical applications	Smoked	(21 CFR-123.3S)
Injected/internal addition	Smoke flavor added	(USDA Policy Memo 058A-4)

Continual research and development allows for improved flavors and increased applications. This allows for smoke flavors to continue to grow in usage for safer foods, increased production, and a cleaner environment.

Food Engineering Applications in Seafood Processing

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Fish oil purification and adsorption technology

Small fish processors and entrepreneurs are interested in producing small-scale, cost-effective fish oil extraction, clarification, and stabilization methods for oil for human consumption. Unpurified oils produced from fish byproducts contain non-triglycerides such as free fatty acids, oxidized components, protein, minerals, and insoluble impurities that reduce quality. These components need to be removed before the oil will be acceptable to many markets. Conventional fish oil refining is achieved through four separate steps: degumming, neutralizing, bleaching, and deodorizing. Phospholipids are removed by degumming. Free fatty acids (FFA) are precipitated as soaps and removed during the neutralization process. Bleaching clays adsorb pigments from oil, and oxidized components can be removed by deodorization. The main disadvantages of conventional methods are high refining losses, additional oxidation, and high energy/processing costs. Adsorption technology can potentially provide a simplified process for refining fish oil for human consumption. This research has shown that an adsorption process used for edible oil purification not only removes non-triglycerides but also is a cost-effective process.

Extending shelf life of fresh fish fillets using edible coating

Fish is an extremely perishable food compared to other fresh commodities and is therefore largely marketed in frozen and processed products. Development of these products from Alaska is hampered by the short life of many types of seafood. Frozen storage is a preservation method used to control or decrease biochemical changes that occur during fish storage. However, frozen storage does not completely slow down undesirable reactions such as lipid oxidation that lead to deterioration of fish quality. Preservative compounds such as phosphates are often added to food products to improve their shelf life, water binding, and frozen stability properties. Phosphates can be used in seafood to enhance the water holding capacity, and to improve cooking yield. Antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) have been commonly used by the food industry to improve product quality during product storage resulting in increased shelf life. Much current research focuses on using natural ingredients such as tocopherols, peptides, or preservatives to meet growing consumers demand for foods devoid of synthetic antioxidants and preservatives. Biodegradable polysaccharides or proteins may be used to coat fish fillets to suppress the quality changes during frozen storage. However, studies on extending the shelf life of fish by use of edible coatings have so far been limited. This research has indicated that some biodegradable edible coatings on frozen fish fillets act as barriers to control moisture transfer and oxygen uptake, thus extending the shelf life of salmon fillets.

Properties of hydrolysates derived from fish processing byproducts

Large amounts of protein-rich byproducts from the seafood industry are discarded or processed into fish meal. Novel processing methods are needed to convert seafood byproducts into more profitable, marketable forms. Many of these protein-rich seafood byproducts have a range of dynamic properties and have the potential to be used in foods as binders, emulsifiers, and gelling agents. Soy and milk proteins are widely used in many sectors of the food industry, while amino acids and peptides are gaining their uses in energy drinks and other beneficial applications. Proteins from fish processing byproducts can be modified to improve their quality and functional characteristics by enzymatic hydrolysis. This research shows how fish protein hydrolysates (FPHs) can be prepared forming new and/or improved properties from the peptides.

Developing microencapsulated fish oil powder for nutraceutical markets

Attempts to incorporate fish oil into food formulations have had limited success because of "fishy" flavors in the finished products. The main problem of food enrichment with omega-3 poly-unsaturated fatty acids (PUFA) is the unpleasant fishy flavor of fish oil that has a negative influence on food acceptability. One of the technologies proposed for protection of fish oil is microencapsulation, defined as a process that makes it possible to transform oil into powder, where the

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small droplets of oil are surrounded by a dry matrix of proteins and/or carbohydrates. Microencapsulated DHA-rich fish oil could fulfill the physical and nutritional requirements of ingredients for enrichment of variety of foods such as milk and bakery products, salad dressings, and juice drinks.

Properties of an engineered fish protein powder

Fish processing byproducts from cold-water marine species are excellent sources of high quality proteins and offer opportunities to use more fish byproducts as protein for food and feed ingredients and industrial applications. Protein-rich seafood byproducts can have a range of dynamic properties and have potential to be used in foods as binders, emulsifiers, and gelling agents.

Rheological properties of food

Rheology is the study of the deformation and flow of matter. Most rheological testing involves applying force to a material and measuring the flow or change in shape. The textural properties that humans perceive when they consume foods are largely rheological in nature, for example, hardness, creaminess, juiciness, smoothness, brittleness, and tenderness. The stability of foods often depends on the rheological characteristics of their components, e.g., emulsions, spreads, and pastes. The flow of foods through pipes or the ease with which they can be packed into containers is largely determined by their rheology.

Knowledge of rheological properties helps solve problems related to the transfer or movement of bulk quantities of liquids. At low temperatures, impurities of crude fish oil tend to precipitate on the walls of pipes. Solid particles in bulk flow can increase the viscosity of the oil, causing increased pressure drop in the pipeline. Due to increased viscosity, oil flow properties exhibit non-Newtonian behavior. Fish oil refinement involves degumming, neutralizing, bleaching, and deodorizing. Impurities, such as free fatty acids, proteins, moisture, pigments, and volatile flavors, are sequentially removed from the oil. Removing impurities may change the flow properties of the oil. Further study of rheological properties of fish oils will help in the design of proper unit operations, production cost, and final quality evaluation.

Thermal properties of food

In general, foods are subjected to variations in temperature during process, transport, and storage. Temperature changes may cause alterations in physical and chemical properties of food components, which influence overall properties of the final product such as taste, appearance, texture, and stability. A better understanding of the influence of temperature on the properties of foods enables food manufacturers to optimize processing conditions and improve product quality. Therefore, it is important for food scientists to have analytical techniques to monitor the changes that occur in foods when their temperature varies. The most commonly occurring phase transitions in foods are melting, crystallization, evaporation, condensation, sublimation, and glass transitions.

A process that absorbs heat is an endothermic process, whereas a process that evolves heat is an exothermic process. The overall properties of foods may be drastically altered when key components undergo phase transitions, so it is important to have analytical techniques for monitoring these processes. These techniques utilize measurements of physical properties of a material that change when a material undergoes a phase transition, e.g., density, rheology, and heat capacity. Use of thermal analysis, thermogravimetry analyzer (TG), and differential scanning calorimetry (DSC) for oil and fat characterization has gained much interest from food industries. These methods require less time and provide precise stability data. TG analysis could be used to determine the quality of fish oils at different refining steps. Compared to older techniques like the active oxygen method (AOM) or the oxygen bomb method, the TG method offers the advantages of smaller sample size required for analysis, precision, and the ability to evaluate the continuous oxidation process. DSC offers a simple means to investigate characteristics of melting and freezing points of fats. The influence of composition of fat, content of water, production materials, aging, and heat treatment on fat and oil quality can be demonstrated on the basis of DSC investigation. DSC has been used to investigate the thermal conductivity and specific heat, melting and crystallization, oil content, wax coating, and phase transition of foods. This research uses DSC and TG machines to study the stability of fish oil and fish meal.

Development and Characterization of Vacuum Packaged Alaska Pink Salmon (*Oncorhynchus gorbuscha*) Jerkies Made with Marinades

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Abstract

Annual Alaska salmon catches are primarily pink salmon volumetrically. A primary avenue is to process pink salmon into canned or retort pouch products. The low ex-vessel price for pink salmon is ideal for the manufacturing of other value-added products. We developed a jerky made from frozen pink salmon fillet blocks that were band-sawed into thin pieces, eliminating the hand slicing process. Flavorings were added using marinade processes where frozen pieces were dipped in cold solutions. Three marinades were tested: 5% salt (S); 5% salt + 15% brown sugar (SS); 5% salt + 15% brown sugar + 2% commercial antioxidant (SSA). After this process, smoke and drying were applied. Effect of marinade composition on product shelf life at 20°C over a period of 60 days was determined. Processing yields were about 26%, regardless of treatment. Jerky contained initially 72% protein, 18% moisture, 4% ash, and 5% lipid with approximately 215 mg per g of oil of omega-3 fatty acids, mainly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Moisture loss in 60 days was 4% with a concomitant increase of total solids, regardless of the type of marinade. Addition of sugar maintained a_w values of 0.62 to 0.69. The maximum aerobic plate count at 60 days was less than 4.5×10^3 colony-forming units per g, indicating a quality product. Thiobarbituric acid reactive substances (TBARS) values were low and did not exceed 2.7 μmol malondialdehyde per kg for all samples, indicating that storage under vacuum prevented oxidation. The use of antioxidant in the marinade provided additional stability to the product with less loss of polyunsaturated and omega-3 fatty acids being observed in SSA samples at 60 days of storage.

Introduction

In 2005 Alaska pink salmon catch totaled about 420,000 t, of which 71% were pink salmon (ADFG 2005). In Alaska, pink salmon is generally processed into cans or frozen into blocks, with modest volumes being sold as frozen fillets (Oliveira et al. 2005). Pink salmon is a rich protein source and a low fat product with lipid content ranging from about 2 to 4%; however, its omega-3 fatty acid content is approximately 32% making it highly nutritious (Oliveira and Bechtel 2005). Nonetheless, the ex-vessel price for pink salmon in 2005 was low at $0.06 per kg, making it ideal for the manufacturing of value-added products such as salmon jerky. Jerky is a dried ready-to-eat product that is traditionally made from beef. The process of making jerky involves cutting the meat into thin strips, marinating them in brine or sugar solution, and then drying either naturally or using a dehydrator. The resulting product is dry, chewy, and leathery and does not break into pieces when bent completely. Beef jerky is a very popular product in the United States and demand has been growing steadily for many years. Processors seeking new markets for salmon could consider tapping the demand for jerkies by manufacturing one made from fish.

Most salmon jerky is made from minced fish that is extruded, smoked, and dried. This product is nutritious but has rubbery texture that is often objectionable. The most desirable jerky is made from whole muscle that retains the natural texture of fish. Traditionally this has been done through a labor intensive process of hand slicing fresh fish fillets into very thin pieces. The pieces are then marinated in salt or sugar solution to remove moisture from the muscle, thus reducing the water activity (Potter and Hotchkiss 1995a). The water activity of the product is further reduced by subsequent smoking and drying steps. Combined, these steps lower the water activity, increase the microbial safety, and enhance the sensory quality of jerky products (Muratore and Licciardello 2005). When coupled with salting, packaging, and chill storage, smoking produces synergistic effects toward microorganisms and increases the shelf life (Muratore and Licciardello 2005). Additionally, a variety of antioxidants can be added to food products to enhance oxidative stability. Some of the most common ones are butylated hydroxy anisol (BHA), butylated hydroxy toluene (BHT), and tert-butylhydroxyquinine (TBHQ) (Du and Ahn 2000, Ho et al. 1995). Alpha tocopherol, a naturally occurring antioxidant, has been successfully used to protect oxymyoglobin against oxidation by quenching free radicals produced during lipid oxidation (Yin et al. 1993). After the jerky is produced, it should be stored in air-tight plastic packaging or jars. The package should be efficient in protecting the product from physical damage, chemical attack, contamination from biological vectors including microorganisms, and deterioration of product by atmospheric oxygen and water vapor (Potter and Hotchkiss 1995b).

Figure 1. Representative steps for the manufacturing of salmon jerky from salmon blocks: (a) Pink salmon block; (b) Frozen salmon strips; (c) Glazing salmon strips; (d) Glazed salmon strips on the smoking cart (5 trays depicted); (e) Smokehouse; (f) Individually vacuum packaged salmon jerky.

The objective of this study was to develop a process to produce jerky made from frozen pink salmon blocks that were band-sawed into thin pieces eliminating the labor intensive hand slicing process. Flavorings were added by glazing the frozen pieces in cold marinades. Glazed pink salmon slices were smoked, dried, and vacuum packaged. Three marinade compositions were tested to determine their effect on shelf life of vacuum packaged salmon jerkies at selected intervals of time over 60 days of storage at 20°C. The variables measured during the study were proximate composition, fatty acid profile, thiobarbituric acid reactive substances (TBARS), water activity (a_W), and microbial load.

Materials and methods

Fish sampling

Fresh whole grade A pink salmon (*Oncorhynchus gorbuscha*) were obtained from a seafood processing plant on Kodiak Island, Alaska, during summer 2005, and immediately transported on ice to the Fishery Industrial Technology Center (FITC) pilot plant. Salmon used in this study were post-rigor and less than 24 hours post-mortem. Seine caught fish, sampled the same day and from the same fishing vessel, were delivered to the seafood processor in a conventional holding tank that used a standard recirculation chilled seawater system. Whole fish weight ranged from 1.5 to 2.5 kg. Fish were immediately eviscerated and hand filleted and frozen into three 2.5 kg blocks using a horizontal plate freezer (Model Freze-Cel 5549, Dole, Lewisburg, Tennessee).

Manufacturing process

Representative steps for the manufacturing of salmon jerky from salmon blocks are depicted in Fig. 1. Frozen pink salmon blocks were cut into thin strips of dimensions (19 cm × 5 cm × 0.5 cm) using a meat saw "Butcher Boy" (Model B 12, Los Angeles, California). Three alternative marinades were prepared using 500 g of filtered water and 500 g of ice as follows: (S) 5% salt; (SS) 5% salt + 15% brown sugar; (SSA) 5% salt + 15% brown sugar + 2% mixture of antioxidant (tocopherols, ascorbic acid, citric acid, polysorbate 80, and potassium sorbate from Kalsec, Kalamazoo, Michigan). Salmon strips were dipped, while frozen, for 30 seconds in one of the three marinades at the ratio of 2.5 kg salmon strips per l L of marinade. The glazed strips were placed orderly in a vertical cart fitted with nine racks (Enviro-Pak, Clackamas, Oregon), and immediately placed in the electronically controlled smokehouse (Model MP500, Enviro-Pak). Smoking was carried out using hickory wood chips (Hardwood Sawdust, Northeastern Product Corp., Warrensburg, New York). Programming consisted of three distinct process stages. The first stage was 30

minutes long to allow the smoldering plate to reach 230°C, the smoking chamber was kept at 32.2°C, and the damper was closed. The second stage, the smoking stage, was carried out with the damper in the open position, the smoldering plate set at 230°C, the velocity of the wood chip feeder set at the medium range, and the smoking chamber kept at a constant temperature of 37.7°C for a total of 8 h. The final stage, the drying stage, was conducted without smoke and for 2.5 h, the damper was set in the open position and the smoking chamber held at a constant temperature of 48.9°C. Samples were cooled to room temperature, and vacuum-packed using Nylon/PE vacuum bags (40 cm × 25 cm; 100 µm film) using an UltraVac (Model UV2100-B, Koch, Kansas City, Missouri). Samples were stored at room temperature in a ventilated area (20°C ± 2°C) for up to 60 days to mimic storage conditions of the product at a conventional food store display. Processing yields were calculated as the difference between the weight of the salmon blocks and the weight of the finished products.

Sampling and statistical analysis

Jerky samples (S, SS, and SSA) were analyzed for initial proximate composition, fatty acids, TBARS values, and water activity at day 0. Every 15 days samples were analyzed for TBARS and water activity values. At 60 days of storage, samples were analyzed for proximate composition, fatty acids, TBARS values, water activity, and microbial content. Analysis of variance was conducted to compare differences among marinade composition (S, SS, and SSA) and storage time (0 and 60 days) using Statistica v. 7.0 (Statsoft, Tulsa, Oklahoma). In cases where significant differences were observed for the main effects or interactions ($P < 0.05$), the Tukey's Honest Significant Differences test was used to investigate specific differences ($P < 0.05$).

Water activity and proximate composition

Water activity was determined in triplicate on pulverized jerky samples (1 g) using a water activity meter (Aqua Lab, Decagon Devices Inc., Pullman, Washington). Samples were pulverized using a high speed tissue homogenizer (Model 31BL91, Waring Commercial, New Hartford, Connecticut). Determination of moisture and ash were conducted in triplicate according to methods 952.08 and 938.08 (Helrich 1990), respectively. Protein was quantified in triplicate using 3 g of homogenized meat for each replicate analysis on a LECO protein analyzer (Model FP2000, LECO, St. Joseph, Michigan). The nitrogen results were multiplied by 6.25 to determine the percent protein found in jerky samples. Lipid extraction was performed in triplicate using an accelerated solvent extraction system (ASE200, Dionex, Sunnyvale, California) as previously described by Oliveira et al. (2006). A quantity of 5 g of homogenized meat was pre-dried in a vacuum oven (Model 1430, VWR Scientific Inc., West Chester, Pennsylvania) operated overnight at approximately 45°C, and then mixed using a mortar and pestle with the Chem Tube hydromatrix (Varian Inc., Palo Alto, California). Extraction was carried out using cells with a 40 ml capacity under pressurized nitrogen atmosphere (10.34 MPa) at a temperature of 80°C, using HPLC grade dichloromethane (VWR International, Brisbane, California), for a total extraction time of 15 min. Additional parameters for this analysis were: fill time 1 min; heat time 5 min; static time 5 min; flush volume 45%; purge time 120 seconds; 2 static cycles. Solvent was evaporated using a TurboVap® LV Evaporator (Caliper LifeSciences, Hopkinton, Massachusetts) operated at 45°C under nitrogen atmosphere. Percent lipids were determined gravimetrically and recovered lipids were combined to a composite sample and immediately suspended in hexane containing 0.01% BHT and stored in 1 ml amber Teflon®-lined screw-cap tubes (Agilent Technologies, Wilmington, Delaware) flushed with nitrogen and stored at −80°C until preparation of methyl esters for fatty acid analysis. Total solids were determined based on the moisture contents and protein, lipid, and ash contents calculated as a percent dry weight.

Microbiological analysis

The jerky samples were evaluated for microbiological content after storage for 60 days. The samples (5 g each) were immersed in 45 ml of 0.1% peptone (Difco Labs, Detroit, Michigan) water in sterile Whirl-Pak bags (Fort Atkinson, Wisconsin) and mixed using a masticator (silver model, IUL Instruments, Barcelona, Spain) for 2 min. Homogenates were serially diluted in peptone water and 0.1 ml spread-plated on duplicate plates containing plate count agar (Difco) plus 0.5% NaCl. The plates were incubated at 35°C for 24-48 hours and the total plate counts calculated (FDA 2001).

TBARS

Thiobarbituric acid reactive substances (TBARS) analysis was conducted using the method described by Lemon (1975). Malondialdehyde (MDA) content in the samples was expressed as values of TBARS in units of µmoles of MDA per 1 kg of tissue.

Preparation of fatty acid methyl esters and GC analysis

Fatty acid methyl esters (FAME) were prepared in duplicate from each of the six composite lipid samples (S day 0, SS day 0, SSA day 0, S day 60, SS day 60, and SSA day 60). Esterification followed the procedure of Maxwell and Marmer (1983) using C23:0 as internal standard. Fatty acid methyl esters were quantified using a gas chromatography (GC) model 6850 (Agilent Technologies, Wilmington, Delaware) fitted with a DB-23 (60 m × 0.25 mm id., 0.25 µm film) capillary column (Agilent) coupled to a flame ionization detector (FID) (Bechtel and Oliveira 2006). Data were collected and analyzed using the GC ChemStation program (Rev.A.08.03; Agilent). Hydrogen was used as carrier gas at linear flow of 1.0 ml per min with an average velocity of 30 cm per second. The initial nominal pressure of the inlet was

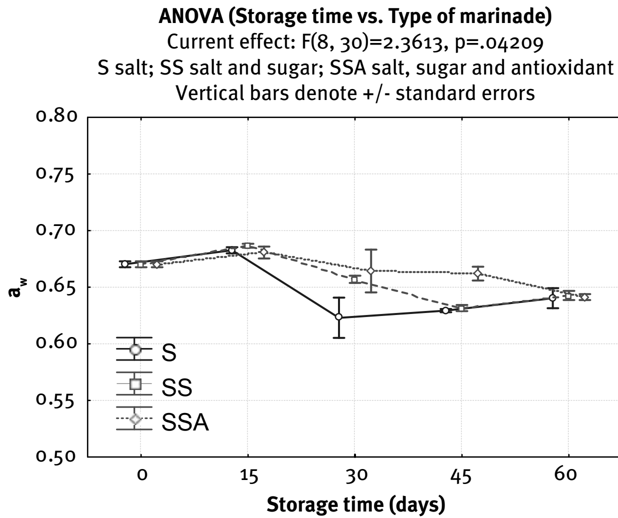

Figure 2. Water activity of vacuum packaged salmon jerkies prepared using three different marinades over 60 days of storage at 20°C.

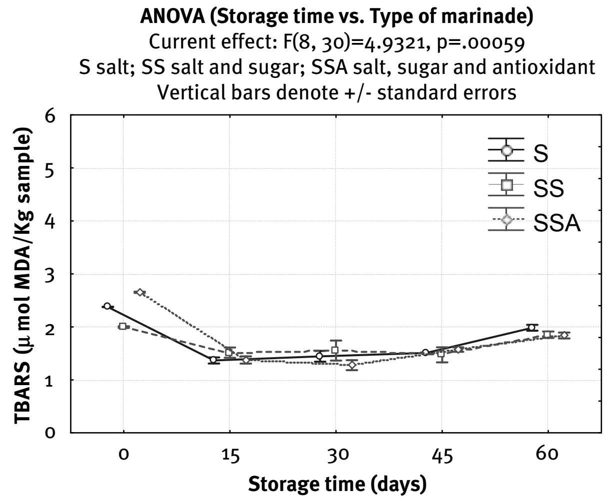

Figure 3. TBARS values of vacuum packaged salmon jerkies prepared using three different marinades over 60 days of storage at 20°C.

105.5 kPa and both injector and detector were held at 275°C. The split ratio was 25:1 and the oven programming was 140°C to 200°C at a rate of 2°C per min, 200°C to 220°C at a rate of 1°C per min, and 220°C to 240°C at a rate of 10°C per min for a total run time of 52 min. The detector was operated at a constant makeup flow of 35 ml per min of nitrogen, with an air and hydrogen flow of 450 ml per min and 40 ml per min, respectively. An autosampler performed the GC injections of standards and sample, and injection volume was 1 µl. The ChemStation enhanced integrator program was used to integrate the chromatogram peaks. All standards used in the identification of peaks were purchased from Supelco® (Bellefonte, Pennsylvania). The standards used were Supelco 37, Bacterial Acid Methyl Esters Mix, Marine Oil #1, and Marine Oil #3.

Results and discussion

Yield and water activity

The total weight of the salmon blocks was 8.8 kg, with an average weight of 2.9 kg per block. After blocks were band-sawed the total weight of the salmon strips was 7.4 kg. Processing yield was approximately 26%, regardless of treatment, yielding a total of 2.3 kg of final product. Fig. 2 depicts the changes in water activity of vacuum packaged salmon jerkies stored at 20°C. Over the 60 days of storage, the a_W was relatively stable, ranging from a maximum of 0.68 (all samples at 15 days of storage) to a minimum of 0.62 (S samples at 30 days of storage). USDA recommends a water activity of <0.80 to prevent growth of *Staphylococcus aureus*, if present, in meat and poultry jerky products (USDA-FSIS 2004). Unlike these products originating from farm animals, fish jerky was not heat-treated to pasteurization temperatures, since other pathogens such as *Salmonella* and *E. coli* O157:H7 are not associated typically with coldwater marine fish. In smoked fish production, contamination by *S. aureus* is prevented by using latex gloves when handling the raw material and product (Himelbloom and Crapo 1998). The initial values of a_W, regardless of type of marinade, were about 0.67, which is below the USDA recommended value for a safe, shelf-stable product.

Proximate composition and microbial content

Table 1 shows the proximate composition and microbial load of salmon jerkies prepared using different marinades at days 0 and 60 of storage under vacuum package conditions at 20°C. Day 0 samples were reported as an average because the composition of the initial product was, as expected, very similar regardless of marinade type. Sugars were not measured because this component of the marinade is an additive only, being deposited as a thin layer on the surface of the jerky strips, and its concentration was considered negligible (<1%) when compared to the quantities of moisture and solids found in the product. A 4% decrease in moisture content occurred during the 60 days of storage, indicating that a moisture-impermeable packaging film is necessary to prevent moisture loss in the jerkies (Table 1). Addition of sugar, used as a humectant for food products, was not effective (at the level added) in preventing moisture loss in the product; S samples showed similar loss as SS and SSA samples. The percent wet weight values for protein, ash, and lipids were converted to percent dry weight, using the total solids content (Table 1). The percent dry weight was the preferred unit for comparison between sampling times (0 and 60 days) because a loss in moisture was observed during storage. There was a small, but significant, reduction in crude

Table 1. Proximate composition and microbial load of vacuum packaged salmon jerkies prepared using three different marinades at 0 and 60 days of storage at 20°C.

	S/SS/SSA	S	SS	SSA
Type of marinade	0 days	60 days	60 days	60 days
Moisture (%)	18.4^a (0.5)	14.4^b (0.3)	14.4^b (0.2)	14.3^b (0.4)
Solids (%)	81.6^b (0.5)	85.6^a (0.3)	85.6^a (0.2)	85.7^a (0.4)
Protein (% dry wt.)	89.3^a (0.6)	84.3^b (0.2)	83.3^{b}c (0.3)	81.9^c (0.2)
Lipids (% dry wt.)	6.2^d (0.1)	9.3^c (0.2)	10.7^b (0.1)	11.9^a (0.2)
Ash (% dry wt.)	4.5^b (0.2)	6.4^a (0.2)	6.0^a (0.1)	6.2^a (0.2)
Microbial load (CFU per g)	NM	3.9×10^3	4.5×10^3	3.9×10^3

S = salt; SS = salt and sugar; SSA = salt, sugar and antioxidant. Different superscript letters within a row signify significant differences ($P < 0.05$). CFU = colony forming units. NM = not measured.

protein content (% dry weight) during the 60 days of storage. Protein degradation through storage was about 5%, 6%, and 7.4% (dry weight) for S, SS, and SSA samples, respectively (Table 1). The relative increase of lipid and ash percent dry weights in salmon jerkies reflected the decrease in moisture and concomitant increase in the total solids content from day 0 to day 60. While the change in ash content was similar to all jerkies, the change in lipids was significantly different. SSA samples showed higher lipid content than SS and S samples after 60 days of storage, and this is relative to the higher protein loss observed in SSA samples. It is possible that addition of antioxidant to the marinade could have led to slightly lower lipid degradation during storage, leading to higher lipid recovery for SSA jerky samples when compared to S and SS samples. Microbial counts were low (<10^4 CFU per g) due to the reduced a_w, and vacuum packaging prevented mold growth.

TBARS values

Fig. 3 shows TBARS values of vacuum packaged salmon jerkies prepared using three different marinades over 60 days of storage at 20°C. Even though significant differences were noted in TBARS during the different sampling times, the variation in TBARS values was small with maximum values not exceeding 2.7 µ moles of MDA per kg sample and minimum values at about 1.2 µ moles of MDA per kg sample. This indicates that little oxidation occurred in the vacuum packaged salmon jerkies through the 60 days of storage, regardless of type of marinade. Similar to our findings, Muratore and Licciardello (2005) reported that TBARS values did not change significantly in vacuum packed liquid smoked swordfish over 42 days of chilled storage (4°C).

Fatty acid profiles

Table 2 depicts the fatty acid profiles of vacuum packaged salmon jerkies prepared using marinades S, SS, and SSA at 0 and 60 days of storage at 20°C. At day 0 the fatty acid profile of S, SS, and SSA were similar, as it can be verified by the small standard errors (Table 2). Therefore, values for all fatty acids at day 0 are reported as an average for all jerkies (S, SS, and SSA). The total amount of saponifiables (sum of all fatty acid methyl esters) present in the extracted oils of salmon jerky at day 0 was approximately 650 g per gram of oil, indicating a good conversion of fatty acids in the form of triacylglycerides and phospholipids to their methyl ester forms. The sum of all fatty acid did not add up to 1 g because of the presence of non-saponifiable material such as glycerol, phosphate, sterols, and free fatty acids. It is noteworthy to point out that the procedure of Maxwell and Marmer (1983) does not esterify any of the free fatty acids possibly present in the extracted lipids. This is the main difference between the KOH/methanol method (Maxwell and Marmer 1983) and the more commonly used BF_3/methanol method (Iverson et al. 1997), which readily esterifies free fatty acids in addition to the fatty acids connected to a glycerol backbone.

The most abundant fatty acids in salmon jerkies at day 0 were DHA (22:6ω3), *cis*-oleic acid (18:1ω9), and palmitic acid (16:0). These three fatty acids made up almost 50% of all fatty acids saponified. These results show that the types of fatty acids that predominate in salmon jerky are beneficial to human health (Gurr 1999), and that the product is a valuable source of omega-3 fatty acids, such as EPA and DHA. Using the 5% (wet weight) value determined for lipid content for jerkies at day 0, the quantity of omega-3 fatty acids in 100 g of product was estimated at approximately 1 g. Concomitantly, the quantity of omega-6 fatty acids in the product at day 0 was very low at 75 mg per 100 g of jerky. Small quantities of *trans*-oleic acid (18:1ω9) were observed in the product at day 0, while *trans*-linoleic acid (18:2ω6) was not detected in any of the chromatograms.

After 60 days of storage, a decrease in the quantities of several fatty acids was observed for S, SS, and SSA (Table 2). Fig. 4 depicts the changes in the fatty acid classes during the 60 days of storage. A decrease in the amount of saponifiables occurred for all samples, indicating lipid degradation, with the S jerky samples showing the highest change, followed by SS and SSA. All fatty acid classes (saturated, monounsaturated, and polyunsaturated) showed a decrease; however,

Table 2. Fatty acid profile of vacuum packaged salmon jerkies prepared using three different marinades at 0 and 60 days of storage at 20°C (mg per g oil).

	Type of marinade			
	S/SS/SSA	S	SS	SSA
	0 days	60 days	60 days	60 days
14:0	24.9 (1.5)	12.6	15.3	15.4
15:0	3.1 (0.2)	1.5	1.9	2.0
16:0	83.1 (3.8)	48.5	48.6	47.9
16:1ω13 and 16:1ω11	2.5 (0.2)	1.4	1.7	1.8
16:1ω9	1.7 (0.1)	0.7	1.0	0.9
16:1ω7	31.6 (1.9)	15.9	17.9	17.2
16:1ω5	1.8 (0.2)	1.0	1.2	1.2
16:2ω4	1.1 (0.1)	1.1	1.1	1.0
17:0	3.2 (0.3)	4.4	3.3	3.0
18:0	23.0 (1.0)	12.4	12.6	12.4
18:1ω9 trans	5.8 (0.6)	4.3	5.7	5.3
18:1ω9 cis	102.5 (8.1)	47.4	54.0	53.0
18:1ω7	16.4 (0.7)	14.8	14.0	12.2
18:1ω5	3.0 (0.3)	2.1	2.7	2.7
18:2ω6 cis	8.9 (0.4)	4.9	6.1	6.6
18:3ω3	6.1 (0.5)	3.2	3.8	4.3
18:4ω3	14.0 (0.9)	9.4	12.7	13.8
20:1ω11	24.4 (2.9)	24.2	33.7	31.7
20:1ω9	15.1 (2.2)	14.7	21.0	19.1
20:1ω7	4.0 (0.5)	1.9	2.1	2.2
20:2ω6	2.7 (0.2)	0.7	1.7	1.9
20:4ω6	3.7 (0.3)	1.6	1.7	1.9
20:4ω3	9.1 (0.7)	4.8	6.6	6.9
20:5ω3	54.4 (2.8)	31.2	35.5	35.9
22:1ω11	44.7 (6.7)	51.7	75.7	71.3
22:1ω9	4.9 (0.7)	5.2	7.7	7.2
22:5ω3	19.8 (1.9)	10.5	13.3	12.6
22:6ω3	112.1 (4.0)	74.3	76.8	84.1
24:1ω9	1.3 (0.2)	5.4	6.5	6.7
Unknown FA	22.0 (2.7)	6.4	9.1	10.3
Saponifiables	650.7 (24.1)	417.9	494.9	492.3
SAT	137.3 (5.7)	79.3	81.7	80.5
MUFA	259.7 (10.4)	190.6	244.8	232.6
PUFA	231.8 (9.4)	141.6	159.2	168.9
ω3	215.5 (8.6)	133.3	148.6	157.6
ω6	15.2 (0.8)	7.2	9.5	10.3

S = salt; SS = salt and sugar; SSA = salt, sugar, and antioxidant. FA fatty acids. Saponifiables = sum of all fatty acid methyl esters. SAT = saturated fatty acids. MUFA = monounsaturated fatty acids. PUFA = polyunsaturated fatty acids.

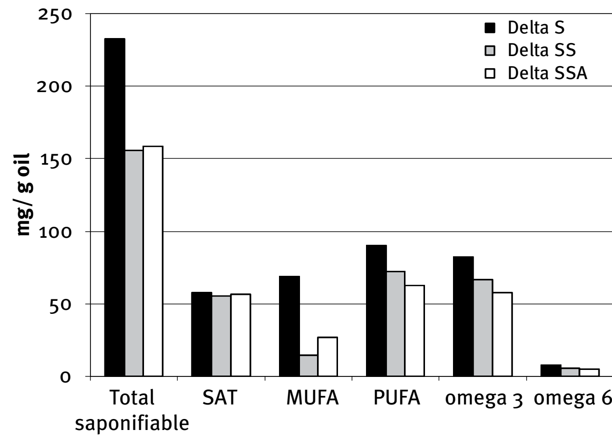

Figure 4. Changes in the fatty acid profile of vacuum packaged salmon jerkies prepared using three different marinades over 60 days of storage at 20°C. (Delta indicates the differences from day 0 to day 60.)

SSA samples tended to retain more polyunsaturated (PUFA) then S and SS samples. This was likely due to the addition of the antioxidant mixture to SSA samples. Various studies on food products indicate that vacuum packaging and modified atmospheric packaging can enhance the shelf life (Ho et al. 1995, Du and Ahn 2000, Lee and Kang 2003, Muratore and Licciardello 2005). Vacuum packaging reduced the oxidative degradation of PUFA in egg yolk when compared to the aerobic pack (Du and Ahn 2000). However, Ho et al. (1995) stated that although vacuum packaging can reduce the oxidation of lipids, for products with high lipid content the addition of an antioxidant should be considered to further protect food products from lipid degradation. In the case of salmon jerky, it seems that the use of the commercial antioxidant mixture in the marinade did not prevent lipid degradation; however, it increased the stability of the lipids during the storage time when compared to S and SS samples (Table 2; Fig. 4). In order to prevent lipid degradation, it may be necessary to increase the thickness of the packaging film in order to reduce oxygen permeability, to reduce light exposure by using opaque packaging, and to increase slightly the quantity of commercial antioxidant in the marinade. Finally, it should be noted that the observed reduction in the amounts of several of the fatty acids did not correlate well with the low TBARS values determined. One possible explanation could be the fact that free fatty acids may have formed by cleavage of the fatty acid moieties of triacylglycerides and phospholipids. As previously stated, free fatty acids were not esterified; thus the fatty acid values reported for the 60 days samples only include those found in the extracted lipids as glyceride and phosphoglyceride moieties.

Conclusion

A process was developed to produce pink salmon jerky from frozen fish blocks that were band-sawed into thin pieces, thus eliminating the hand slicing process. It was possible to add flavorings to the product with a glazing step using a marinade. Results showed that salmon jerky is a good source of protein, and a valuable source of omega-3 fatty acids. However, the 4% moisture loss observed during the 60 days of storage was undesirable, and the use of a moisture-impermeable packaging film will be necessary. The use of antioxidant in the marinade provided additional stability to the product with less loss of polyunsaturated and omega-3 fatty acids being observed at 60 days of storage. Nevertheless, it was noted that preventing lipid degradation is needed to improve product stability, and this may be accomplished by increasing the thickness of the packaging film, by using opaque packaging, and by increasing slightly the quantity of commercial antioxidant in the marinade.

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Using Sustainable Packaging Technologies to Respond to Consumer, Retailer, and Seafood Industry Needs

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Abstract

Although seafood consumption increased from 15.2 pounds per capita in 2000 to 16.5 in 2006, smoked fish remained stagnant (National Fisheries Institute, pers. comm., 2007; Johnson 2007) throughout this time period. Meeting consumer, retailer, and industry needs better could spawn growth in the smoked fish industry. More sustainable practices and packaging can be used to address consumer, retailer, and industry needs while maintaining product quality and safety. This includes sustainable practices and use of emerging packaging technologies.

Triple bottom line (TBL) opportunities for sustainable packaging in the context of consumers, retailers, and manufacturers are considered and explored for applications to the smoked fish consumer, retailer, and industry. Several initiatives, such as the Sustainability Packaging Coalition and global retailer and governmental initiatives that are defining sustainability, are discussed. Packaging technologies of edible/degradable films/coatings and nanopolymer barrier technology are explored in the context of attaining more sustainable packaging.

The consumer, retail, and seafood industry needs can be addressed while maintaining the distinct characteristics of smoked fish using packaging technologies.

Introduction

Sustainable packaging has been applied to the smoked fish industry for centuries. Long ago, consumers eating smoked fish needed to responsibly dispose of the leaf package that fish smokers used to wrap the fish. There has always been a need to lessen the impact of packaging on the environment. However, since the mid 1970s, in response to consumer need for convenience, our increased access to global markets, and the multitude of new packaging material technologies, we have fundamentally altered the way we package products such as smoked fish. This has created a crisis.

The crisis is the access to and then disposal of the materials—metal, glass, plastics, wood—that we use for packaging. The crisis manifests itself in the generation of greenhouse gases and the depletion of natural resources. In the 1990s, the United States (primarily California) started laws and regulations related to packaging and, in the EU and BRIC countries (Brazil, Russia, India, China), a solutions-based framework began taking place led by Germany's Green Dot program. Now, in the twenty-first century, the packaging industry has moved globally toward the development of an initiative called "more sustainable packaging." Sustainable initiatives have reached the forefront of global concerns with former U.S. Vice President Al Gore and the Alliance for Climate Protection being the co-recipients of the Nobel Peace Prize in 2007. Global industries, such as the smoked fish industry, face sustainable packaging challenges due to the shelf life, package requirements, and food safety concerns in a globally competitive market. Suppliers are developing increasingly more sustainable solutions to lessen the impact of packaging while meeting food industry, consumer, and retailer needs.

Sustainable packaging defined

Sustainable packaging does not have a succinct definition, but the SPC (Sustainable Packaging Coalition, http://www.sustainablepackaging.org), an international corporate consortium formed in 2005, has a widely accepted definition, which is that sustainable packaging:

1. Is beneficial, safe, and healthy for individuals and communities throughout its life cycle;
2. Meets market criteria for performance and cost;
3. Is sourced, manufactured, transported, and recycled using renewable energy;
4. Maximizes the use of renewable or recycled source materials;
5. Is manufactured using clean production technologies and best practices;
6. Is made from materials healthy in all probable end of life scenarios;
7. Is physically designed to optimize materials and energy; and
8. Is effectively recovered and utilized in biological and/or industrial cradle-to-cradle cycles.

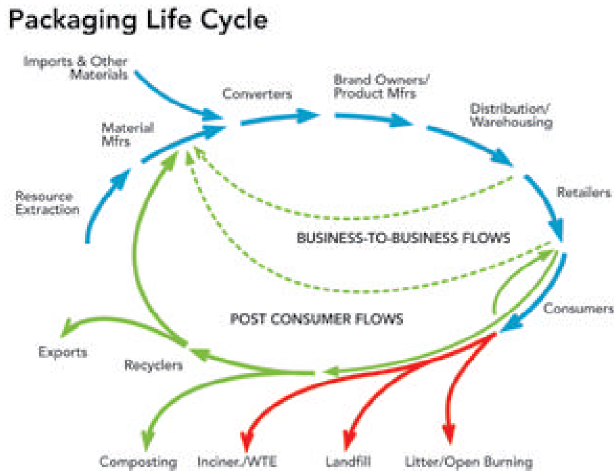

Figure 1. Packaging life cycle. © GreenBlue 2006.

Membership in the SPC reflects the commitment of major industry players to more sustainable packaging. Over 70 major retailers, packaging manufacturers, and product manufacturers have joined the SPC. As part of its approach to support innovation and effective new technologies and share best practices, SPC has conducted studies to use in outreach initiatives. These studies focus on renewable energy, recycled content, design for recycling, and source reduction.

Further, to evaluate the impact of a package change, the SPC has licensed a screening tool from Environmental Defense called MERGE™, which calculates a profile of package design based on seven criteria that can provide the user with feedback on the package's sustainability.

Cradle to cradle

Packaging sustainability now moves toward a cradle-to-cradle philosophy that all things are renewable and there should be no waste of energy. This encompasses biological recovery (managed composting), technical recovery, such as recycling and reuse, and energy recovery, to recover value from spent package materials. The packaging life cycle considers the life of a package from cradle to cradle.

Life cycle analysis (LCA) and carbon footprinting quantify the impact of the entire packaging life cycle from material origin in the earth to disposal and recovery as noted in Fig. 1. Zero carbon generation is an ultimate goal in the packaging life cycle. In fact, Norway's plan to achieve zero carbon status by 2050 provides concrete direction of how the packaging industry will use resources in the future (Sand 2007).

Sustainable packaging drivers

Motivation for pursuing more sustainable packaging aligns with many corporate philosophies that now embrace the 3Ps—profit, people, planet. This is known as the triple bottom line (TBL). With TBL opportunities, profits can be generated by doing what benefits people and minimizes planetary impact. As the packaging industry realizes that sustainability can be profitable because materials and energy costs are conserved, industry leaders are, in some cases, ahead of pending global regulations. Three TBL areas of opportunity for manufacturers and retailers in attaining more sustainable packaging are

1. Sourcing more sustainable materials. For example, more sustainable paper/paperboard/corrugated from forests certified by the Program for the Endorsement of Forest Certification schemes (PEFC), Sustainable Forestry Initiative (SFI), or the Forest Stewardship Council (FSC) can be selected.
2. Altering energy usage. Reducing dependence on coal and oil for energy through the increased use of alternative energies such as water, wind, and solar energy is more sustainable and often more profitable. The EU plan is to generate 22% of energy from renewable resources by 2010.
3. Reducing emissions into air, land, and water, and reducing emissions in industries provides opportunities for less costly disposal.

Packaging manufacturers drive sustainable packaging

Selecting one material versus another in an effort to attain more sustainable packaging involves use of the LCA information. The Toxics in Packaging Clearing House (www.toxicsinpackaging.org) offers a guide to assess packaging materials. Deciding which material will result in more TBL benefits requires package specific LCA studies. For example, since polyethylene (PE) is made from the less renewable resource (oil) than paper (made from wood), PE is often perceived as less sustainable than paper. However, PE actually requires less energy and water to make from oil, and PE generates less emissions than paper during production and can be recycled indefinitely unlike paper fibers.

Efforts from packaging manufacturers have focused on achieving TBL benefits through energy reduction. Large companies such as Alcoa have made major inroads to making production more sustainable. Smaller companies have made impacts too, such as Curtis Packaging, which generates 100% of its electricity from wind and hydroelectric power.

Food manufacturers drive sustainable packaging

Food companies have remained dedicated to the 3 Rs (reduce, reuse, recycle) concept and using the packaging life cycle to understand opportunities. In fact, the U.S. recycling rate has doubled to 32% over the last 15 years, accomplished through a systems approach to packaging. For example, labels were used to support the thinned sidewalls of plastic cups, layers

of recycled plastics were used in bottles, and some bottles were redesigned so that they could be sanitized, refilled, and returned to distribution. In terms of reducing the amount of material, the United States has seen

1. 2 liter polyethyleneterephthalate (PET) bottles are 25% lighter now than in 1977.
2. Glass jars are 43% lighter now than they were in 1970.
3. Four cans can be made from one pound of aluminum, compared to 22 pounds in 1972.

Packaging food safely needs to be a primary concern in the use of more sustainable food packaging. Primary food packaging cannot always comply with environmental initiatives because of food safety concerns, oxygen barrier performance, strength needs, etc. While opportunities to reduce, reuse, and recycle primary food packaging are limited, increasingly applications are becoming more viable. For example

1. Reduce primary packaging. The Dasani water bottle redesign reduced PET use by 10,000 metric tons in 2005.
2. Reuse primary packaging. The Hillshire Farm Deli Select line of pre-sliced lunch meat uses Glad Ware® containers that can be washed and reused.
3. Recycling primary packaging. The UK will open the first food grade PET recycling plant in late 2007.

So, the food industry has focused on sustainable initiatives with its secondary and tertiary packaging that does not have direct product contact. Reuse and recycling of corrugated is viable since corrugated transports 70% of the world's liquid and solid materials from producer to retailer/consumer. For example, pallets and individual bulk containers can be designed for reuse in closed loop environments in which the pallets are returned to the initial user, or non-closed loop in which pallets and shipping containers have multiple purposes by different manufacturers, using incoming containers as padding for products. For example, the change to a 64 oz rectangular bottle for juice saves Ocean Spray Cranberries, Middleboro, Massachusetts, 11% in shipping-related corrugated costs and reduces energy expenses throughout distribution and logistics.

Retailers drive sustainable packaging

Retailers use TBL to maintain their competitive advantage. For example, Marks and Spencer has reduced CO2 emissions by 30% from 2003 to 2006 and by 2012 plans are to reduce their carbon footprint by 80%. Worldwide, other retailers have followed these actions to achieve more TBL profits. The world's largest retailer driver, Wal-Mart, introduced in the fall of 2006 the Wal-Mart scorecard to enable measuring its 60,000 suppliers on sustainability criteria. The program begins in 2008 with the goal of reducing overall packaging by 5%. Wal-Mart expects to save $3.4 billion from this initiative. In the late fall of 2007 the company will introduce a packaging scorecard on private label suppliers so that their buyers will have all the information on packaging alternatives and sustainable packaging. And FreshDirect has launched environmentally friendly initiatives in an effort to increase the sustainability of the source, packaging, and distribution of seafood (Progressive Grocer 2007). Regulations and retailer driven packaging restrictions relating to sustainability and consumer perception of "overpackaging" can also be addressed with new package solutions (Greenberg 2005, Gulbrandsen 2005).

Consumers drive sustainable packaging

Consumers are driving sustainable packaging as well. The LOHAS (lifestyles of healthy and sustainable) segment of the U.S. population is 30% (2007). These consumers demand more sustainable packaging. EPA figures for 2005 show that 31% of the material that municipalities dispose of is packaging. Smoked fish packaging (and all food packaging) is highly visible to consumers. Consumers, on the average in the United States, dispose of 3 pounds of packaging waste per day. No wonder consumers, especially the LOHAS consumer, consider packaging a target for social responsibility (Sand 2007).

Global laws and regulations drive sustainable packaging

In the early 1990s, governments did make strides in advocating more sustainable packaging under the theme of the 3 Rs. The U.S. Congress deliberated the concerns of the public. Rather scary at the time was the naive congressional solution to put all foods in PET because it is easily recycled. This was never legislated.

Now, Europe is a major driver of sustainable packaging. The EU Directive on Packaging Waste (94/62/EC) has been replaced by 2004/12/EC. The new directive requires

1. 60% by weight of all packaged wastes must be recovered or incinerated as waste;
2. Increased numbers of incineration plants with energy recovery;
3. Established targets of 55-80% packaging waste by weight to be recycled; and
4. The National Packaging Waste Database is to be developed by 2008.

The United States is the only industrialized country to not have national environmental packaging legislation. The U.S. and countries predicted for high growth such as Mexico, Indonesia, Vietnam, Korea, and Turkey are predicted to be pulled along to comply with the EU regulations.

Technology enables more safe and sustainable food packaging

The food industry has a unique opportunity to employ new packaging technologies to meet the need for more sustainable packaging. Risk assessments demonstrate the need to control microbial growth (WHO 2004). Applicable technologies are edible film (with or without antimicrobials), degradable packaging, use of alternative fiber materials, modified atmosphere packaging (MAP), and use of nanocomposites (polymers bonded with nanoparticles and montmorillonite, a clay material).

Antimicrobial polymers

Antimicrobial polymers have been employed in the food industry to retard the growth of microorganisms that pose a food safety threat. Antimicrobial edible film technology is seamless to the consumer as it effectively dissolves into food prior to consumer purchase (Brody 2005). This technology has been employed worldwide to enable food to have a longer shelf life, reduce food safety concerns related to large distribution networks, increase energy efficiency, and improve product confidence. Edible and antimicrobial packaging can also reduce the amount of synthetic packaging needed to package a given product.

Chicken, for example, is coated with an edible antimicrobial film made of methyl cellulose with nisin—an antimicrobial agent to retard the growth of *Salmonella*. Silver zeolite in plastic film has been shown to retard microbial growth (McKinley 2003). The International Smoked Fish Conference Proceedings reports on the use of nisin for Listeria monocytogenes control in cold-smoked salmon (Neetoo et al. 2008). Recent research reported at IFT 2007 demonstrated that smoked salmon coated with a whey protein film containing lysozyme (an enzyme with antimicrobial properties extracted from egg white) is effective against *Listeria monocytogenes* (Min et al. 2007). This kind of laboratory research assists the seafood industry in enhancing its food safety record and enabling more sustainable packaging solutions.

Degradable polymers

A more sustainable future is coming with degradable polymers like polycaprolactam (PCL), polybutylene succinate (PBS), polyhydroxybutyrate-hydroxyvalerate (PHbV), polyhydroxyalkanoate (PHA), and polylactic acid (PLA) made from more renewable sources. PLA is compostable under specific conditions, not degradable. PHbV has been used extensively in Japan and the EU since the mid 1990s for shampoo bottles and food service applications but not for prolonged contact with food. The multitude of degradable polymers reaching the marketplace warrants a review of the appropriate technology as well as further research in the area of product safety, degradable polymers, and potential shelf life extension with the use of degradable polymers. LCA studies on alternate material applications are essential to consider the environmental impact of, for example, energy required to grow and process the crops into polymers and the impact of degradation.

Alternatives to wood derived materials

The smoked fish industry (just like the entire food industry) has an opportunity to reduce the environmental impact of secondary and tertiary packaging through the use of non-wood fibers. While wood fibers are renewable, there are fibers that can be made into paper/paperboard/corrugated that offer more TBL opportunities. There are numerous alternative fibers that can be used and that have been traditionally been used in various regions of the world. Alternative fibers include kenaf, bagasse, jute, cotton, flax, switchgrass, and hemp. Fiber selection is dependent on the climate and growing season. However, genetic engineering research may soon enable alternative fibers to grow in more locations throughout the world (Sand 2007).

Kenaf, a hibiscus species, is widely thought to hold the most promise as a wood alternative. Kenaf, a seasonal product that grows 10-14 feet tall, offers a distinct advantage over hardwood and softwood trees because its long fibers contribute strength to paper/paperboard/corrugated. Containers made from kenaf then require less material, can be made reusable, and are more recyclable. This is because the fiber length allows for recycling 10 times versus the 3-5 times for containers made from wood fibers. Kenaf applications in the packaging industry have been in blending polyethylene (PE) with up to 85% kenaf for reusable composite crates, pallets, and board (Sand, 2007)

Numerous alternative fibers are being used. For example, Wal-Mart sells kiwi packaged in an Earthcycle® palm fiber molded tray wrapped with compostable NatureFlex® film and marked with a compostable label and water-based inks (E-Wire 2006).

Nanocomposites

Nanocomposites improve the barrier and mechanical properties packaging with nano (10^{-9}) size entities within a polymer structure (Lagaron et al. 2005). This is the new opportunity in one of the 3Rs—reduce. Nanotechnologies are predicted to grow from a $66 million business in 2003 to $360 million by 2008 (Downing-Perrault 2005). This growth will result in the use of less packaging and will play an important role in improving shelf life for products such as smoked fish.

Modified atmosphere packaging

While modified atmosphere packaging (MAP) technology has been employed for decades, new computer modeling capability allows food industry professionals to evaluate product shelf life as a function of altering the material and gases used to package the product. This is a tremendous aid

to evaluate new technologies in packaging related to more sustainable packaging.

Summary

New approaches to achieving the 3Rs—including LCAs, the cradle-to-cradle concept, and the use of global regulations—provide an opportunity to create more sustainable primary, secondary, and tertiary food packaging. A TBL focus provides motivation for pursuing more sustainable packaging in the smoked fish industry. Technologies are available to meet consumer, retailer, and industry needs for more sustainable packaging.

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Use of Antimicrobial-Coated Plastic Films to Control *Listeria monocytogenes* on Cold-Smoked Salmon

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Abstract

Cold-smoked salmon (CSS) is an important value-added, ready-to-eat food. Unfortunately, CSS has been classified a high-risk product for *Listeria monocytogenes* contamination. The objective of this study was therefore to examine if antimicrobial films could be used as a hurdle to inhibit *L. monocytogenes* on the surface of this food. Low density polyethylene (LDPE) films incorporating nisin at a level of 2,000 IU per cm^2 wrapped on CSS inoculated with 5×10^2 CFU per cm^2 of *L. monocytogenes* reduced the counts by 3.9 log CFU per cm^2 (compared with control) after 56 (4°C) and 49 (10°C) days of storage while reductions of 2.4 and 0.7 log CFU per cm^2 were achieved for samples inoculated with a high level of *L. monocytogenes* (5×10^5 CFU per cm^2) after 58 (4°C) and 43 (10°C) days, respectively. For samples packaged in film coated with 500 IU per cm^2 of nisin, reductions ranged between 0.5 and 1.8 log CFU per cm^2. LDPE films incorporating nisin (500 IU per cm^2) in combination with various organic salts—sodium lactate, sodium benzoate (SB), sodium diacetate, and potassium sorbate (PS)—were subsequently assessed for their antilisterial efficacy on CSS. Binary combinations of nisin with 0.3% of PS or 0.1% of SB appeared to be most effective against *L. monocytogenes*. LDPE films incorporating nisin/0.3% of PS and nisin/0.3%PS/0.1%SB were found to be highly antilisterial and demonstrated comparable effectiveness reducing the populations of *L. monocytogenes* by a maximum of 4.2 log CFU per cm^2 (relative to the control) after 10 weeks of refrigerated storage.

Introduction

Listeria monocytogenes has long been established as an important food-borne pathogen. However, the incidence of multistate food-related listeriosis outbreaks has increased dramatically recently and *L. monocytogenes* is thus considered a pathogen of major concern (Ryser and Marth 2007). The organism is ubiquitous in the environment, has been detected in many different foods, can grow aerobically or anaerobically at temperatures as low as 1°C, and the minimum infectious dose is not known (Pelroy et al. 1994). However it is assumed that less than 1,000 organisms could cause infection depending on susceptibility of the victim (FDA 2002). *L. monocytogenes* is particularly a major concern for several high-risk subpopulations, such as the elderly, pregnant women and their fetuses, neonates, and those who are immuno-compromised (CFSAN 2001).

In the United States, the organism has been found in a variety of fishery products, both raw and ready-to-eat (Weagant et al. 1988, Buchanan et al. 1989, Noah et al. 1991), including cold-smoked fish. As a facultatively anaerobic and psychrotrophic bacterium, *L. monocytogenes* can grow in ready-to-eat foods which are vacuum-packed and cold-stored, such as slightly salted, cold-smoked, hot-smoked, sous-vide, or other minimally processed fish products. The consumption of fish or seafood has been associated with human listeriosis outbreaks (Lennon et al. 1984, Facinelli et al. 1989, Brett et al.1998). In lightly preserved fish products, the prevalence of *L. monocytogenes* varied from 0 to 75% being highest in cold-smoked fish (Embarek 1994). In a Finnish study, 15% of 40 cold-smoked salmon (CSS) samples collected from retail trade, marketplaces, and fish plants contained *L. monocytogenes* at levels of 1.0-4.3 log CFU per g (Keto and Rahkio 1998). Previous reports have shown that growth of *L. monocytogenes* can occur in artificially contaminated CSS when it is stored at refrigeration temperatures of 4 and 10°C for prolonged periods of up to 30 days (Farber 1991, Guyer and Jemmi 1991). Recalls of contaminated fishery products have resulted in severe economic losses to producers in both domestic and international markets.

Processing of CSS includes no recognized critical control point for *L. monocytogenes*, and this product probably cannot be produced completely free of this pathogen (Gram 2001). Since it is virtually impossible to totally avoid the contamination of lightly preserved fish products with *L. monocytogenes* and emerging technologies such as high pressure processing, irradiation and pulsed-electric field are not feasible for use on packaged CSS, interest in the incorporation of generally-recognized-as-safe (GRAS) biological (e.g., bacteriocins such as nisin) and chemical (e.g., sodium lactate [SL], sodium diacetate [SD], sodium benzoate [SB], and potassium sorbate [PS]) antimicrobial compounds into packaging material has been renewed. Nisin is a natural antimicrobial polypeptide that has been shown to inhibit *L. monocytogenes*. Several studies have reported the effectiveness of nisin in delaying and reducing growth of *Listeria* spp. in model systems (Jamuna et al. 2005, Boziaris and Nychas 2006) and in RTE products (Jamuna et al. 2005, Geornaras et al. 2006) including smoked salmon (Zuckerman and Ben Avraham 2002,

Al-Holy et al. 2004). Sodium lactate (SL) is used to enhance flavor, control microbial growth, and increase shelf-life for meat and poultry products (Duxbury 1990, Lamkey et al. 1991, Papadopoulos et al. 1991), and also delay growth and toxin production by *Clostridium botulinum* in cooked poultry and fish products (Anders et al. 1989, Maas et al. 1989). Inhibition of *L. monocytogenes* by SL has been demonstrated in a variety of products including comminuted chicken and beef model systems, cook-in-bag roasts (Shelef and Yang 1991,Unda et al. 1991, Chen and Shelef 1992) and comminuted salmon model system for cold-process salmon (Pelroy et al. 1994). Currently, SL is allowed at 4.8% for the decontamination of seafood products (Code of Federal Regulations title 21, section 181.23). Sodium diacetate (SD) is approved as a GRAS substance for miscellaneous and general-purpose usage (Code of Federal Regulations title 21, section 181.23). The maximum acceptable daily intake of SD for humans is 0 to 15 mg per kg body weight (Doores 1993). It is used in baked goods because of its inhibitory activity against bread mold and rope-forming bacteria such as *Bacillus subtilis*. Previous studies have already proven the antilisterial efficacy of SD in model systems (Shelef and Addala 1994), in turkey slurries (Schlyter et al. 1993), on wieners and cooked bratwurst (Glass et al. 2002), as well as in CSS (Vogel et al. 2006). Other chemical antimicrobial agents that have also been considered as potential ingredients to inhibit the growth of *L. monocytogenes* were sodium benzoate (SB) and potassium sorbate (PS). Both are GRAS additives and have been shown to inhibit growth of gram-positive bacterial pathogens such as *L. monocytogenes* in media, as well as in and on meat systems (Robach and Sofos 1982; El-Shenawy and Marth 1988; Wederquist et al. 1994; Samelis et al. 2001; Islam et al. 2002a,b).

The objectives of this research were to (i) evaluate the potential of nisin-coated plastic films to control the growth of *L. monocytogenes* on vacuum-packaged CSS and (ii) to investigate the possible synergistic antilisterial effects of nisin and organic salts when co-incorporated into films for application onto CSS.

Materials and methods

Effect of nisin-coated plastic films on L. monocytogenes growth on CSS

Experimental design

A 3 × 3 × 2 factorial design was used for collecting data totaling 18 treatments. Three nisin concentrations (0, 500, and 2,000 IU per cm^2), three inoculum levels (0, 5 × 10^2, and 5 × 10^5 CFU per cm^2 of salmon surface) and two storage temperatures (4 and 10°C) were investigated during each trial.

Preparation of nisin-coated plastic films

A coating solution was prepared by mixing 1.4 g of methylcellulose (MC) (Sigma-Aldrich, St. Louis, Missouri) and 0.6 g of hydroxypropyl methylcellulose (HPMC) (Sigma-Aldrich) in 40 ml of 95% ethanol (Fisher Scientific, Hampton, New Hampshire) until completely dissolved, followed by the addition of 40 ml of sterile distilled water. Subsequently, 1.2 ml of polyethylene glycol 400 (Fisher Scientific), a plasticizer, was added to the mixture. Once a homogeneous mixture was obtained, 1 g (for film containing 500 IU of nisin per cm^2 film surface) or 4 g (for film containing 2,000 IU of nisin per cm^2 film surface), nisin (Sigma-Aldrich) was dissolved in 20 ml of 0.02 M acetic acid (Fisher Scientific) and the resulting nisin solution was well mixed with the coating liquid mixture (Franklin et al. 2004, Cooksey 2005). Nisin-coated films were prepared by taping low density polyethylene (LDPE) films (DuPont Company, Wilmington, Delaware) to 20 × 20 cm glass plates, and the coating solution was cast onto the films using a thin-layer chromatography plate coater (TLC, CAMAG, Muttenz, Switzerland). The coated films were then air-dried at room temperature overnight.

Bacterial strains and culture conditions

Three most nisin-resistant *L. monocytogenes* strains, PSU1 (Serotype 1/2a), PSU2 (Serotype 1/2a), and PSU21 (Serotype 4b), were used in this part of the study. The strains were maintained on tryptic soy agar plus 0.6% yeast extract (TSAYE) (Difco Laboratories, Sparks, Maryland) plates and stored at 4°C. Each strain was grown separately in tryptic soy broth plus yeast extract (TSBYE) for 24 h at 37°C and 100 ml of each overnight culture was transferred to fresh TSBYE broths for another 24 h incubation. On the day of the experiment, a 1 ml volume of each culture was combined to provide a three-strain mixture and then readjusted with 0.1% peptone water to cell densities of ca. 10^8 and 10^5 CFU per ml, which served as the inoculum. Serial dilutions were plated onto TSAYE plates and incubated at 37°C for 24 h to determine cell numbers.

Inoculation of CSS samples and packaging

Freshly processed CSS (*Salmo salar*) samples were obtained from a producer. They were kept frozen at −20°C and thawed at 2 ± 2°C (<4°C) for 1 day immediately before use as described by Besse et al. (2004). Slices of CSS were punched aseptically into 5.7 cm diameter round pieces weighing 10 ± 1 g. The samples were surface-inoculated with 125 μl of appropriate dilution of the three-strain cocktail of *L. monocytogenes* to achieve final concentrations of 5 × 10^2 or 5 × 10^5 CFU per cm^2 of salmon surface. Control and nisin-coated (500 and 2,000 IU per cm^2) LDPE films were wrapped around the inoculated and un-inoculated CSS samples. The wrapped samples were then inserted into 3 mm thick high barrier pouches (nylon/polyethylene, Koch Supplies, Kansas City, Missouri) and subsequently sealed using a vacuum-packaging machine (Model Ultravac 225 with digital control panel, Koch Equipment, Kansas City). The samples were stored at either 4°C for 56 days or 10°C for 49 days. Counts of *L. monocytogenes* were determined at selected time intervals.

Microbial enumeration of inoculated samples

For microbiological analysis, CSS samples were individually placed in stomacher bags that contained 40 ml of 0.1% ster-

Table 1. Formulations for the various ternary combinations of antimicrobials incorporated into films.

Nisin (IU per cm²)	PS (% g per cm²)	SB (% g per cm²)
0	0.0	0.0
500	0.0	0.0
500	0.0	0.05
500	0.0	0.1
500	0.15	0.0
500	0.15	0.05
500	0.15	0.1
500	0.3	0.0
500	0.3	0.05
500	0.3	0.1

PS = potassium sorbate; SB = sodium benzoate.

Table 2. Combinations of antimicrobials incorporated in films used in the long-term refrigerated storage study.

Nisin (IU per cm²)	PS (% g per cm²)	SB (% g per cm²)
0	0	0
500	0.3	0
500	0.3	0.1

PS = potassium sorbate; SB = sodium benzoate.

ile peptone water and stomached for 2 min. Serial dilutions were made in 0.1% peptone water, and counts of *L. monocytogenes* were determined by an overlay method (Kang and Fung 1999). Briefly, the serial dilutions were spread plated on solidified TSAYE agar plates and the plates were incubated at 37°C for 3 h. Approximately 7 ml of modified Oxford medium (Difco Laboratories) at 45°C was overlaid on the TSAYE plates. The plates were incubated at 37°C for 48 h and black colonies on the plates were counted. Occasionally, colonies were confirmed to be *L. monocytogenes* using a BAX™ for Screening/*Listeria monocytogenes* PCR assay (Qualicon-DuPont, Wilmington, Delaware). The absence of *L. monocytogenes* in the CSS samples was confirmed by a primary enrichment in UVM broth (Difco Laboratories) and a secondary enrichment in Fraser broth (Difco Laboratories) according to the USDA Microbiology Laboratory Guidebook (USDA 2005).

Effect of plastic films incorporating nisin and organic salts on L. monocytogenes growth on CSS

Experimental design

Since plastic film containing 500 IU per cm² of nisin alone was not very effective against *L. monocytogenes* on CSS and it is a relatively expensive antimicrobial, four GRAS preservatives were used in combination with 500 IU per cm² of nisin to determine whether the effectiveness of the antimicrobial films could be enhanced. The efficacy of nisin alone (500 IU per cm²) and in binary combination with four organic salts (SL, SD, SB, and PS) was initially screened during storage at ambient temperature (22°C) to select the two most effective organic salts. The four organic salts and their concentrations were SL (0.3%), SD (0.25%), SB (0.1%), and PS (0.3%). The concentrations of the organic salts were calculated on a weight/film surface area ratio. Once the two chemical preservatives with the highest efficacy were selected, ternary combinations (Table 1) incorporating nisin with these preservatives at low and high concentrations were then evaluated during storage at ambient temperature. Finally, the two most effective combinations (Table 2) were chosen and their antimicrobial efficacy with respect to *L. monocytogenes* on CSS was studied during refrigerated storage (4°C) of the samples over 10 weeks.

Preparation of antimicrobial-coated plastic films incorporating nisin and salts of organic acids

The nisin-containing solution was prepared as described previously and supplemented with SD, PS, SB, or 60% SL commercial syrup and subsequently mixed well (Franklin et al. 2004, Cooksey 2005). The coating solution was degassed by placing it in the vacuum-packaging machine. Antimicrobial films were prepared for the following study by casting the coating solutions onto LDPE plastic films using the TLC plate coater.

Inoculation of CSS samples and packaging

A cocktail of five strains of *L. monocytogenes* was used to increase genetic variability and these included two nisin-resistant strains, PSU1 and PSU21, and three other strains, PSU9 (Serotype 1/2b), F5069 (Serotype 4b), and Scott A (Serotype 4b). The cultures of *L. monocytogenes* were prepared and mixed to form a cocktail as described above. CSS samples were inoculated with the cocktail to a final level of 10^3 (for the extended refrigeration study) or 10^5 (for the ambient temperature screening study) CFU per cm². The samples were wrapped in control (plain LDPE film) or antimicrobial films prepared above. They were then inserted into the high barrier pouches and vacuum-packaged. Packages were stored at room temperature (22°C) for 10 days for the screening study and stored at 4°C for 10 weeks for the final refrigerated storage study. Storage at 22°C was chosen to accelerate the selection of the most effective compounds against *L. monocytogenes* prior to an extended storage at refrigeration temperature. Counts of *L. monocytogenes* were determined at selected time intervals.

Statistical analysis

Three independent trials were conducted for each experiment. Single samples were serially diluted and plated in duplicate at each sampling time. Colony counts were converted to log CFU per cm² and means and standard devi-

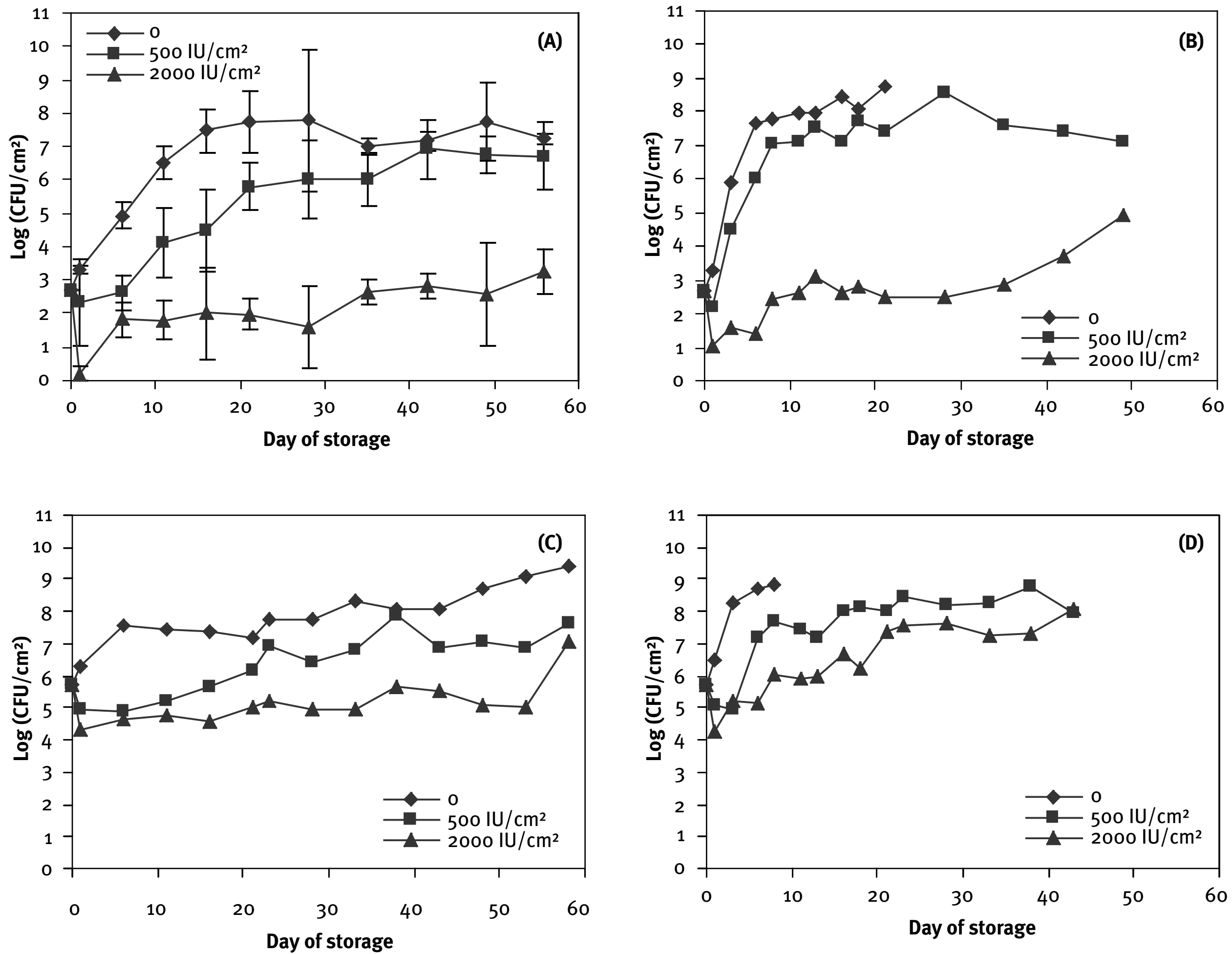

Figure 1. Effect of nisin concentration on low density polyethylene (LDPE) film, inoculation level, and storage temperature on the growth of *L. monocytogenes* on cold-smoked salmon. Samples were inoculated with low (5×10^2 CFU per cm²) or high (5×10^5 CFU per cm²) levels of *L. monocytogenes* and stored at 4 or 10°C. (A) low inoculation level and 4°C; (B) low inoculation level and 10°C; (C) high inoculation level and 4°C; and (D) high inoculation level and 10°C. Error bars represent ± 1 standard deviation. To make the figures easy to read, error bars are only shown for (A).

ations were calculated. Data were analyzed using Microsoft Excel. Differences in mean log CFU per cm² among treatments to determine statistically significant differences ($P < 0.05$) of mean values between treated and control samples and between treatments at each sampling time were calculated using the Tukey-Kramer test at $\alpha = 0.05$ level.

Results

Effect of nisin-coated plastic films on the growth of L. monocytogenes on CSS

Samples of CSS obtained from the producer had no detectable *L. monocytogenes* before inoculation. Counts of *L. monocytogenes* on CSS samples packaged in plain and nisin-coated LDPE films and stored at 4 or 10°C are shown in Fig. 1. LDPE film (no nisin) allowed *L. monocytogenes* on CSS to grow rapidly, especially at higher storage temperatures. The populations of *L. monocytogenes* reached >7.0 log CFU per cm² after 16 days storage at 4°C and 6 days of storage at 10°C, respectively, for the low inoculation level. For the high inoculation level, counts reached >7.0 log after 6 days storage at 4°C and 3 days storage at 10°C. Nisin-coated films slowed down or inhibited the growth of *L. monocytogenes*. The overall trend for both inoculation levels and storage temperatures showed that counts for samples packaged in film coated with 2,000 IU per cm² of nisin were consistently lower than those for samples packaged in film coated with 500 IU per cm² of nisin which in turn were lower than those for samples packaged in control film.

For salmon samples inoculated at 500 CFU per cm² level and stored at 4°C (Fig. 1A), LDPE film coated with 2,000 IU per cm² of nisin significantly inhibited the growth of *L. monocytogenes* on the surface of CSS ($P < 0.05$) through 56 days of storage compared with the control film without nisin.

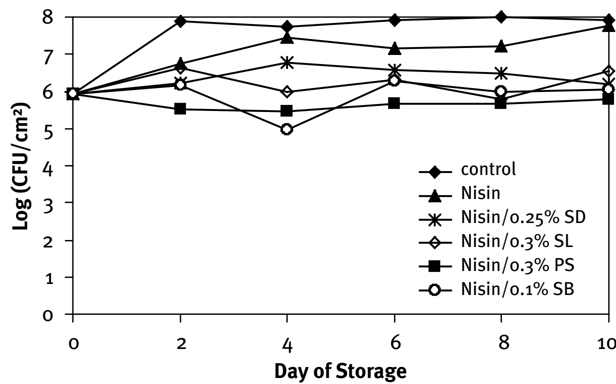

Figure 2. Populations of *L. monocytogenes* on vacuum-packaged cold smoked salmon (CSS) slices packaged with antimicrobial films incorporating generally regarded as safe (GRAS) preservatives and storage at ambient temperature for 10 days. The average standard deviation was 0.5 log CFU per cm². PS = potassium sorbate; SB = sodium benzoate; SL = sodium lactate; SD = sodium diacetate.

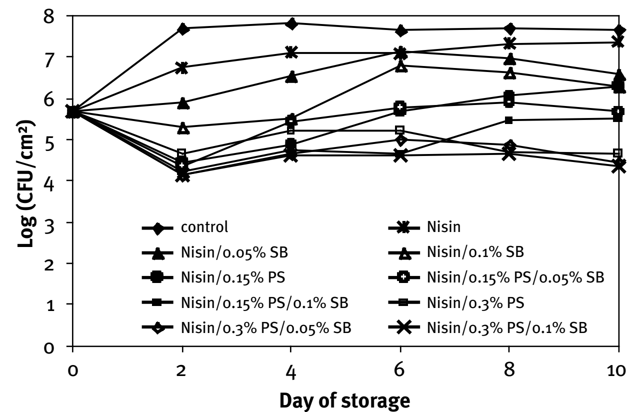

Figure 3. Populations of *L. monocytogenes* on vacuum-packaged CSS slices packaged with antimicrobial films incorporating nisin (500 IU per cm²) with low or high concentrations of potassium sorbate (PS) and/or sodium benzoate (SB) and storage at ambient temperature for 10 days. The average standard deviation was 0.5 log CFU per cm².

Films coated with 500 IU per cm² of nisin allowed *L. monocytogenes* to grow although growth was slower than samples wrapped in plain films. Similarly, for CSS samples inoculated at 500 CFU per cm² level and stored at 10°C (Fig. 1B), films containing 2,000 IU per cm² of nisin significantly suppressed growth of *L. monocytogenes* throughout the 49 day-storage compared with control films. Although the counts for the samples packaged in films coated with 500 IU per cm² of nisin were consistently lower than the control samples, the degree of inhibition was marginal. These results indicated the importance of nisin concentration for inhibiting *L. monocytogenes* on the surface of CSS.

For CSS samples inoculated at 5 × 10⁵ CFU per cm² and stored at 4°C (Fig. 1C), film incorporating 2,000 IU per cm² of nisin significantly inhibited growth of *L. monocytogenes* when compared to the control samples ($P < 0.05$) and achieved a reduction of 2.4 log CFU/cm²(compared to the control) by the end of the trial. Film incorporating 500 IU per cm² nisin also hindered the growth of *L. monocytogenes* on salmon although this film was not as effective as films coated with 2,000 IU per cm². For samples inoculated at a level of 5 × 10⁵ CFU per cm² and stored at 10°C (Fig. 1D), nisin (500 or 2,000 IU per cm²) did not significantly inhibit any growth of *L. monocytogenes* on salmon ($P > 0.05$) although counts were consistently lower for nisin-treated samples relative to control samples throughout the 43-day study.

Effect of plastic films incorporating nisin and organic salts on L. monocytogenes growth on CSS

Screening of binary combinations of antimicrobials

Representative samples of CSS had no detectable *L. monocytogenes* before inoculation. Counts of *L. monocytogenes* for inoculated CSS slices are represented in Fig. 2. The initial concentration of *L. monocytogenes* on inoculated CSS was approximately 6.0 log CFU per cm². At day 2, the control group had higher counts than all other treatments with nisin/0.1%SB (6.0 log) and nisin/0.3%PS (5.8 log) samples having significantly lower counts relative to the control ($P > 0.05$). By day 10, *L. monocytogenes* on the control groups had grown steadily reaching a maximum of 8.0 log while binary combinations incorporating nisin with 0.1% of SB or 0.3% of PS had produced the lowest counts compared to all other formulations. Although the differences were not significant, the population densities at day 10 for nisin/0.3%PS and nisin/0.1%SB film-applied samples were more than 1.8 log units lower than those of the control group.

Screening of ternary combinations of antimicrobial treatments on CSS

Based on the growth trend of *L. monocytogenes* and the mean population density at day 10, treatments with PS and SB were selected for the second stage of the experiment. In this phase of the study, these treatments were each utilized at 0, 0.15, 0.3% and 0, 0.05 or 0.1% for PS and SB respectively in a 3 × 3 factorial design (Table 1). Fig. 3 shows the effects of the treatments on the growth of *L. monocytogenes* on CSS. The initial counts of *L. monocytogenes* on CSS was about 5.6 log CFU per cm² and grew steadily over the 10-day period reaching a maximum count of 7.6 log. This demonstrates the ability of the pathogen to undergo abundant growth on vacuum-packaged CSS without antimicrobials during storage at ambient temperature. Over the storage period, the counts for all other treatments were consistently lower relative to the control groups. Ternary combinations incorporating nisin with SB and PS at both low and high concentrations significantly ($P < 0.05$) inhibited the growth of *L. monocytogenes* achieving a

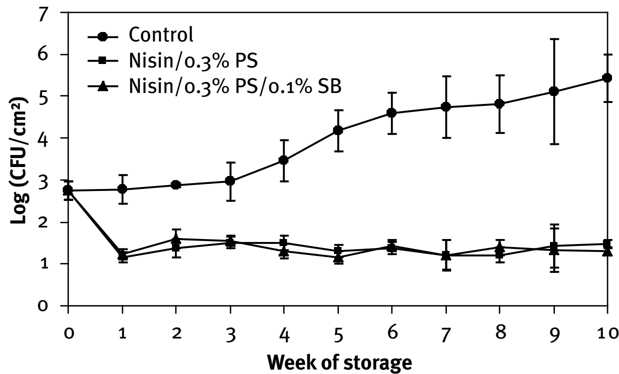

Figure 4. Effect of plastic films coated with nisin/0.3%PS and nisin/0.3%PS/0.1%SB on the growth of *L. monocytogenes* on cold smoked salmon (CSS) during storage at 4°C for 10 weeks. Error bars represent ± 1 standard deviation. PS = potassium sorbate; SB = sodium benzoate.

reduction of 2.0 to 3.3 log relative to the control. The ternary combination with the highest degree of inhibition consisted of nisin with 0.3% of PS and 0.1% of SB.

Evaluation of long-term antilisterial effectiveness of selected antimicrobial combinations on CSS

In the last phase of this study, the organic salt treatment groups which were most effective against *L. monocytogenes* were then selected in the investigation of their long-term antilisterial effectiveness in CSS. The two treatment groups chosen for this study were nisin/0.3%PS and nisin/0.3%PS/0.1%SB (Table 2). The latter formulations were shown to be most effective with no significant difference between them. Nisin/0.3%PS was investigated in the long-term experiment alongside nisin/0.3%PS/0.1%SB as there is a concern that high levels of chemical preservatives required to inhibit the growth of *L. monocytogenes* can have a negative impact on sensory properties and may discourage customers from repurchase. In addition, use of minimal concentrations of antimicrobial ingredients may provide safer and more acceptable options for manufacturing without adversely affecting product quality. Based on these reasons, these two antimicrobial formulations were chosen for the refrigerated storage study.

Un-inoculated samples were free of *Listeria* spp.; therefore, all *Listeria* spp. found in the inoculated samples originated from the inoculum and was *L. monocytogenes*. The mean population of *L. monocytogenes* on inoculated treated slices as recovered just after inoculation was 2.7 log CFU per cm² (Fig. 4). *L. monocytogenes* populations grew in CSS samples with no treatment after storage for 10 weeks at 4°C reaching approximately 5.4 log. Packaging with antimicrobial films brought about a significant reduction of the population of *L. monocytogenes* after 10 weeks achieving reductions of 4.0-4.1 log (relative to the control) by the end of the study. Binary and ternary combinations of nisin (500 IU per cm²) in the presence of 0.3% of PS were found to be equally effective against *L. monocytogenes* ($P > 0.05$).

Discussion

Effect of nisin-coated plastic films on L. monocytogenes growth on CSS

In this study, the effect of storage temperature, nisin concentration on LDPE film, and inoculation level on the growth and survival of *L. monocytogenes* was investigated. At 4°C (low and high inoculum levels) and 10°C (low inoculum level), it was found that the degree of inactivation or growth inhibition of *L. monocytogenes* was directly related to the concentration of nisin; however, for samples inoculated with high levels of *L. monocytogenes* and stored at the abusive temperature of 10°C, neither nisin concentration was adequate in controlling proliferation of the pathogen. Moreover, storage at 4°C allowed slower growth of *L. monocytogenes* than at 10°C regardless of the inoculation level or the nisin concentration on the films. Therefore the combination of refrigeration temperature (4°C) storage in the presence of packaging film incorporating 2,000 IU per cm² nisin was found to be the most effective of the treatment conditions for limiting the growth of *L. monocytogenes* on CSS. The fact that nisin delayed the growth of *L. monocytogenes* populations in CSS at both low and high inoculum levels show that nisin can be used to effectively control post-processing contamination of *L. monocytogenes* in CSS.

Based on the results of this study, it can be concluded that *L. monocytogenes* survival on CSS was dependent on nisin dose, and it was inhibited remarkably well in the presence of film incorporating nisin at a level of 2,000 IU per cm². Results from this study also show that the application of nisin film in packaging CSS has the potential to overcome the problems associated with the post-process contamination of this food by *L. monocytogenes*.

Effect of films incorporating nisin and organic salts on L. monocytogenes growth on CSS

Although nisin exhibited marked listeriostatic activity at a level of 2,000 IU per cm², it is an expensive substance and hence the cost of commercial use of this agent may be prohibitive for the food industry, especially when high concentrations are needed to achieve satisfactory antimicrobial effects. Typically it was common to use only one chemical antimicrobial agent in a food product for preservation purpose (Busta and Foegeding 1983). However, in recent years, the use of combined agents in a single food system has become more frequent. The use of combined antimicrobial agents theoretically provides a greater spectrum of activity, with increased antimicrobial action against pathogenic organisms. It is believed that the combined agents would act on different metabolic elements within similar species or strains which would theoretically result in improved microbial control over the use of one antimicrobial agent alone (Santiesteban-Lopez et al. 2007).

In this study, it was demonstrated that the antilisterial effect of low level of nisin could be greatly enhanced by the co-incorporation of chemical preservatives in films. The type of preservative and its concentration were the main factors that affected the growth of *L. monocytogenes* at ambient temperature. Of all the salts of organic acids tested in this study, 0.3% of PS and 0.1% of SB demonstrated the most consistent antilisterial activity in the ambient temperature screening study. Indeed, the listeriostatic ability of SB and PS on their own has been reported elsewhere on numerous occasions (Hiromi et al. 2007). Moreover, the synergistic antilisterial effects of nisin in the presence of PS or SB have also been demonstrated. Fang et al. (1997) tested varying amounts of nisin in combination with either PS or SB for effectiveness in inhibiting growth of *Staphylococcus aureus* C10 and *Bacillus cereus* B7 inoculated on vacuum-packaged and non-vacuum-packaged vegetarian food. Data indicated that during the 14-day storage at 4°C, vacuum-packaged samples treated with 5×10^3 IU per g nisin and 0.12% SB significantly ($P < 0.05$) decreased the counts of *S. aureus* C10 and *B. cereus* B7 by 2.6 and 3.0 log CFU per g, respectively. Buncic et al. (1995) reported strong listeriocidal effects when 0.3% of PS was used in combination with other antimicrobial agents such as nisin (400 IU per ml). The combination of sorbate and nisin caused a 4.5 log reduction of the population of *L. monocytogenes* inoculated into a buffered culture broth (pH 5.5, 4°C) in 5 weeks.

SB and PS were subsequently combined with nisin in ternary combinations incorporating the salts at low or high concentrations. Preliminary screenings of these combinations at ambient temperature identified three formulations: nisin/0.3%PS, nisin/0.3%PS/0.05%SB and nisin/0.3%PS/0.1%SB as having the greatest inhibitory impact on *L. monocytogenes* by the end of the accelerated shelf-life study. Inhibition and inactivation of *L. monocytogenes* in the presence of nisin, PS and SB was affected by the concentration of PS and SB (i.e., more rapid at higher than lower concentrations). Previous studies have indeed shown that SB/PS combinations can exhibit strong listeriostatic and listeriocidal abilities. Glass et al. (2006) evaluated the control of *L. monocytogenes* on ready-to-eat uncured turkey and cured pork-beef bologna with combinations of benzoate, propionate, and sorbate. Their studies confirmed that sorbate and benzoate mixtures can delay or prevent the growth of *L. monocytogenes* in processed meats compared with similar formulations without these agents.

Finally, nisin/0.3%PS and nisin/0.3%PS/0.1%SB were selected and compared for their effectiveness during an extended storage period at refrigeration temperature. The 10-week refrigerated storage study results indicated that antimicrobial packaging incorporating a low concentration of nisin and 0.3% of PS (with or without SB) applied onto the surfaces of CSS inhibited the growth of *L. monocytogenes*. Nisin/0.3%PS and nisin/0.3%PS/0.1%SB reduced the populations of *L. monocytogenes* relative to the control samples by 4.1 log. These combinations exhibited comparable antimicrobial effectiveness when incorporated in films with no significant difference in antimicrobial activity ($P > 0.05$). Therefore, it was concluded that LDPE films coated with 500 IU per cm^2 of nisin and 0.3% of PS could represent a feasible option to control *L. monocytogenes* on CSS.

Temperature was also recognized as one of the main extrinsic factors affecting *L. monocytogenes* growth. As expected, growth was more rapid at 22°C than at 4°C. At 4°C, *L. monocytogenes* grew, but its lag phase was much longer than at 22°C. The ability of *L. monocytogenes* to grow in CSS at 4°C was not unexpected. This bacterium is known to grow at refrigeration temperatures. The growth of *L. monocytogenes* at 4°C was reported by Barbosa et al. (1994) who studied the behavior of 125 strains of *L. monocytogenes* in a synthetic medium. Higher storage temperature promoted faster growth of *L. monocytogenes* than lower temperature storage; however, the effect of low temperature alone was minimal in retarding the growth of *L. monocytogenes* compared with the more extensive growth-repressive effect of a preservative.

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Globalization and Aquaculture: Challenges, Opportunities, and Questions for the Smoked Seafood Industry

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Introduction

The world seafood industry is changing rapidly. This paper describes some of the most important changes that are happening and suggests questions people in the smoked seafood industry should be thinking about in order to respond to these changes.

Globalization of the world seafood industry

The world economy is experiencing far-reaching changes that are collectively referred to as "globalization." Among the causes and consequences of globalization are increasingly reliance on markets; reductions in trade barriers and expansion of trade; world economic integration in markets for resources, goods, services, labor, and capital; movement of production to low-cost producers; consolidation and integration resulting in larger and more powerful firms operating in many countries; technological revolutions in communications and transportation; growing consumer incomes in developed and developing countries; and increasing consumer expectations for lower prices, convenience, variety, and quality.

Globalization is rapidly transforming seafood production, processing, distribution, and retailing. Globalization is bringing

- Rapid expansion of seafood trade.
- Rapid growth in aquaculture.
- Rapid technological change in processing, transportation, logistics, and marketing.
- The emergence of major new seafood markets as countries in East Asia and formerly socialist countries industrialize and become more open to trade.
- Increasing consolidation and growing concentration of market power in the retail and food service industries.
- Growing demand for new seafood product forms.
- Increasing pressure on seafood suppliers to improve quality and lower costs.
- Restructuring of seafood distribution networks.
- Shift in labor-intensive seafood processing to countries with low labor costs.
- Stricter international standards for food handling and safety.
- Emerging demands for environmentally sustainable and socially responsible seafood production.

Globalization is contributing to the growth of two kinds of seafood buyers—very different in scale—who represent two kinds of market opportunities for smoked seafood producers. Ever-larger retail and food service chains are emerging as a result of consolidation in the retail and food service industries—companies such as Wal-Mart, Costco, Safeway, and Darden Seafoods (which operates numerous restaurant chains) (Fig. 1). These companies represent a growing share of the seafood market in the United States, Europe, and Japan—as well as in new markets such as Russia and China. For these companies, a single person may make the seafood buying decisions for hundreds of stores and restaurants. In general, to meet the needs of their companies and their customers, these buyers want to buy seafood products that

- Can be supplied consistently, reliably, and in large volumes.
- Can be supplied at stable and competitive prices.
- Are viewed by consumers as safe, convenient, and attractive.
- Are traceable through the entire chain of production and distribution.
- Are perceived by consumers as safe and healthy.

In general, it is difficult for small seafood producers to sell to these large buyers, because they cannot consistently and reliably supply large volumes.

Globalization is also contributing to the growth of "specialty" stores and restaurants catering to consumers who are relatively less concerned about price and relatively more interested in quality, variety, and other product attributes. Buyers for these stores and restaurants are more likely to be looking for seafood products that consumers will perceive as high quality, healthy, organic, natural, local, and/or environmentally and socially responsible.

"Specialty" stores and restaurants, ranging from local businesses to larger chains such as Whole Foods, represent a market opportunity for smaller seafood producers who

Figure 1. **Large chains such as Wal-Mart and Costco keep costs low by buying in very large volumes. These companies want to buy products that can be supplied consistently in large volumes at stable and competitive prices.**

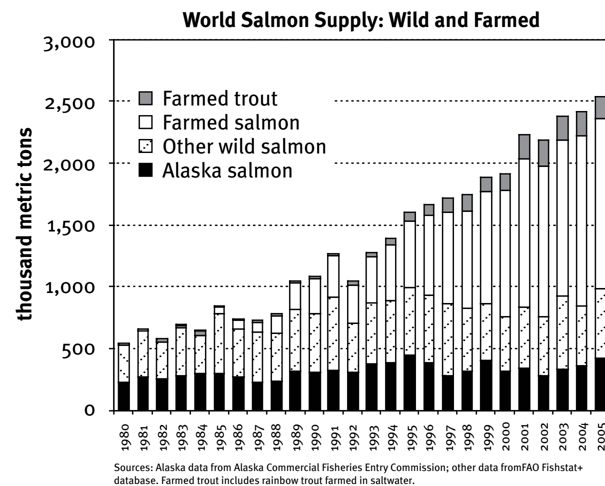

Figure 2. **Between 1980 and 2004, Alaska wild salmon's share of world salmon supply fell from 42% to 17% as farmed salmon production grew dramatically.**

are not able to meet the volume demands of large buyers. However, these smaller buyers also want products that can be supplied reliably and at competitive prices.

What do seafood buyers want? What do consumers want? There isn't any single answer. There are many market niches with many kinds of buyers selling to many kinds of consumers who want many kinds of products—all with different requirements for volume, price, quality, and other product characteristics. The important point for smoked seafood producers is to have a clear understanding of the needs of the buyers and consumers of their products—and to recognize that globalization is bringing all buyers and consumers more options for meeting those needs, and making all buyers more interested in seafood quality, safety, and traceability.

The aquaculture revolution

An aquaculture revolution is occurring in world seafood production. Farmed seafood accounts for a large and growing share of world seafood production (Fig. 2). Fish farmers now provide most of the supply of four of the top six seafood species consumed in the United States: shrimp, salmon, catfish, and tilapia.

Aquaculture is growing rapidly because it can meet market demands—particularly those of large retail and food-service buyers—for predictable, year-round, and growing supply of good quality seafood (Fig. 4). It is critical for wild seafood producers to recognize the reality of the market advantages farmed fish have in meeting market demands, particularly those of large buyers. It is critical for wild seafood producers to work to overcome—to the extent possible—the market disadvantages created by varying and uncertain seasonal supply and inconsistent quality.

Understanding how wild and farmed fish compete requires looking at the entire systems for producing, processing, distributing, and marketing wild and farmed fish. For example, the relatively higher costs of growing salmon rather than catching them wild are significantly offset by the lower costs of processing farmed salmon in facilities that operate year-round.

Some wild seafood producers point to challenges faced by aquaculture, such as diseases, negative environmental

Figure 3. Fresh tilapia for sale at Swanson's Store, Bethel, Alaska, April 2002.

effects, limits to the supply of wild fish–based feeds, and dependence on artificial coloring. While these are real challenges for fish farmers, they do not mean that aquaculture will not continue to grow in supply or that competition from aquaculture will go away.

Certainly fish farming has limited potential in some places, and farmed fish products have limited potential in some markets. But globally there is enormous potential for future growth in aquaculture. Fish farmers can respond to challenges they face by changing what fish they grow and how and where they grow them. For example, salmon farmers can substitute vegetable-based feeds for fish-based feeds. The rapid growth in consumption of farmed salmon, shrimp, tilapia, and other products proves that buyers and consumers will accept farmed products (Fig. 3).

Wild seafood producers can continue to compete successfully as world aquaculture production grows. As the supply of fish grows—as fish becomes more widely available in more places and more product forms over more of the year—consumer demand for fish also expands. As consumer demand expands, more market niches are created for wild seafood—including smoked wild seafood products (Fig. 5).

Questions for the smoked seafood industry

How can people in the smoked seafood industry best prepare for and respond to the changes in the seafood industry that globalization and aquaculture are bringing? A starting point is thinking about answers to questions you are likely to face, such as the following:

How will your competition change? Globalization is opening new markets for you around the world—but it is also opening new markets for new competitors. What new competitors may emerge for you? Where will they get their fish? What technologies will they use? What competitive advantages and disadvantages will they have compared with you? What will you need to do to compete successfully?

What new safety and health concerns are likely to arise for smoked seafood? Global seafood consumers are becoming increasingly safety and health conscious. With globalization of the press, concerns about a product's safety and health effects—including concerns that have no basis in fact—can be reported almost instantaneously worldwide, with immediate and dramatic effects on buyer and consumer demand. Groups that promote or claim to promote consumer health and food safety—both government and private—may intensify the media focus on particular concerns. Anyone in the food industry should think carefully about what real or perceived concerns might arise with respect to the safety and healthfulness of their products, and how to minimize the risks associated with such concerns.

What regulatory changes may occur for smoked seafood products? Reflecting in part concerns about food safety and health, regulatory agencies worldwide are adapting stricter regulations on processing technology, smoking temperatures and time, packaging, labeling, and product naming. What regulatory changes may affect your products? How would you respond?

What kinds of traceability and sustainability will markets demand for the fish you use to make your products? Increasingly, buyers for your products are likely to want to know where your fish or shellfish were harvested, and whether they were harvested from "sustainable" fisheries. Can you trace the origin of your fish? How will you assure buyers that they were "sustainably" harvested?

How may demand change for your products? If you make a traditional smoked seafood product and sell it to a traditional market for that product, how is that market likely to change? What are the demographics of the consumers of your products? If they are aging, are younger consumers likely to continue to demand the product?

What are potential new markets and products for smoked seafood? How can you reach these markets? How can you work together with others in your industry to encourage growth of new markets?

Conclusions

World seafood markets are experiencing rapid and profound change. Globalization is creating new opportunities and new

Figure 4. **A sculptor for the annual Anchorage "Salmon on Parade" competition—like many Alaska fishermen—thinks farmed salmon are "insipid, toxic, dyed, and mushy." But market reality is that farmed salmon meets the needs of large buyers for a high-quality product available fresh in large volumes consistently throughout the year.**

Figure 5. **Wild and farmed smoked salmon. Aquaculture represents a major new source of supply for the smoked seafood industry, and new competition for wild seafood producers.**

challenges for seafood producers. Aquaculture is profoundly changing who is producing seafood, what is produced, and what it costs to produce it. For seafood producers, these changes are bringing more opportunities in more markets around the world, more competition from more suppliers around the world, and more demands from buyers. The producers who understand and prepare for the changes that are happening are most likely to succeed.

In understanding and preparing for change, two common-sense strategies—which apply to any industry at any time—are becoming increasingly relevant for smoked seafood producers:

- Learn about your markets and your competitors. Travel. Attend trade shows. Meet your customers—the people you sell to, the people they sell to, and the people who ultimately consume your product. Learn what they want. Learn what they think about your product, and how it compares with your competitors' products. Learn how they think the market is changing, and why. Think about your competitors. Who are they? Where are they? What advantages and disadvantages do they have compared with you? What can you learn from them?
- Work together as an industry to address issues of common interest. Smoked seafood producers have many areas of common interest. You can confront new challenges and take advantages of new opportunities more effectively if you work together with others in your industry. Some of the areas in which the smoked seafood industry may most benefit from cooperation include generic marketing, marketing research, and monitoring and lobbying for regulatory changes.

Fishing for Answers? Consult the Internet!

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During my first two decades in Cooperative Extension work with the University of California, responding to industry inquiries sometimes required that I walk across the UC Davis campus to Shields Library and hike up two flights of stairs and trudge into the book stacks to delve for answers to food processing and regulatory inquiries. Another laborious and time-consuming option was to drive to the UC Davis School of Medicine library, which was located even farther from my office. There was no easy access to electronic databases and online resources when I launched my career in public service. Thus a significant amount of time and effort was spent physically going to the campus libraries and manually reviewing the U.S. Code of Federal Regulations, Food Science and Technology Abstracts and other key resources.

Then in the 1990s, widespread public access to the Internet helped revolutionize the way our extension program conducts outreach and how we deliver information to people. Also in 1995, the Hazard Analysis Critical Control Point (HACCP) food safety management system regulation for domestic seafood processors and importers was enacted by the U.S. Food and Drug Administration. Unlike other regulations, effective implementation of HACCP to control hazards (biological, chemical, and physical) is scientifically dependent. HACCP training or experience is federally mandated for processors and importers. The seafood industry (processors and inspectors) has questions about HACCP and they rely on Sea Grant extension programs and the Internet for answers.

This presentation gives a brief overview on search engines, provides criteria to help you determine the credibility of the Web sites you are accessing, presents information on online discussion groups, and includes key Internet resources that are helpful to seafood processors.

Using search engines

In 2006 the Pew Internet and American Life Project published the results of its telephone survey, "The Internet as a Resource for News and Information about Science" (http://www.pewinternet.org/pdfs/PIP_Exploratorium_Science.pdf). Of the 2,000 adults surveyed, 1,447 used the Internet. Of those who use the Internet, 87% use it as a tool for getting scientific information. The most popular tool for searching is via a search engine. Using a search engine, key words (search terms) are entered and the "go" or "enter" icon should be clicked, which then links the viewer to a variety of Web pages on the Internet topic being searched by the engine database within seconds.

The University of California Berkeley library offers an online tutorial, "Finding information on the Internet: A Tutorial" (http://www.lib.berkeley.edu/TeachingLib/Guides/Internet/FindInfo.html) which highly recommends using Google.com as a search engine, and for a second opinion it recommends Yahoo! Search and Ask.com (http://www.lib.berkeley.edu/find/types/websites.html).

To assist in demonstrating the utility of the search engines, I used two different terms (*Listeria* and smoked seafood, and *Listeria* in smoked fish) to conduct a search via http://www.google.com, http://www.ask.com, and http://www.yahoo.com.

The results vary depending on the kind of search terms that you use (Table 1.)

Results from the search engines provide links that guide the viewer to Web pages containing the information that is available on the Internet.

If you visit a Web site that does not have its own search engine posted, you can use an existing search engine (i.e., Google.com, etc.) to comb through information on that particular Web site. Using the search box of the search engine site, type [search term] site: [Web address]. If "www" is part of the Web address, omit it and type in the rest of the address. Instead of www.fda.gov, use fda.gov. So, a search for "*listeria*" on the Seafood Network Information Center, and the Food and Drug Administration Web sites would respectively look like:

Listeria site:seafood.ucdavis.edu
Listeria site:fda.gov

The search results will roughly give you the pages as if you were using a search engine that is incorporated on the Web site.

There are some disadvantages in searching for information on the Internet via a search engine:

- The site may no longer be accessible (the server may no longer be online, is temporarily unavailable, or the Web page has been altered or removed).

Table 1. Results of search for *Listeria* and smoked seafood and fish terms among search engines (October 7, 2007).

	Search terms and number of results	
Search engine Web sites	*Listeria* in smoked seafood	*Listeria* in smoked fish
Google.com	49,100	86,700
Ask.com	22,300	34,800
Yahoo.com	4,200	6,040

- You may have to use the search engine on the specific Web site that you are visiting. The search engines may not penetrate proprietary databases.

The advantages of searching for information on the Internet include:

- Availability 24 hours a day, 7 days a week.
- Convenience (no more treks to the campus library!).
- Much of the information is free, but you'll need to know how to discern the credibility of the site. This is discussed next.
- Real-time access worldwide gives you immediate information as it occurs.

One very convenient tool to use in searching through long documents is the "edit" function in the browser. Click "edit" in the top left corner of your browser, then "find in this page" and you can key in a search term that quickly scans through a document to the search term and stops at the search term. For example, it could be challenging looking for information related to smoked seafood in the HACCP regulation; it is several pages long (http://www.cfsan.fda.gov/~lrd/searule3.html). By using the edit function to search for a term such as "smoked," within a fraction of a second you'll be brought to the smoked information that is located in the regulation. This method saves a lot of time and is a big convenience!

What I like about using the Internet as a sourcing tool for information is that I usually do not have to leave my desk to find scientific documentation. Having the computer on your desk with a link to the Internet is like having a mini-library at your disposal. The Internet is a powerful and invaluable information-sourcing tool for you to have on the job.

How do you determine if a Web site is credible?

There's a lot of good information on the Internet, but there is also a lot of misinformation. The UC Berkeley Library also gives advice on criteria to consider in evaluating the credibility of an unfamiliar Web site (http://www.lib.berkeley.edu/TeachingLib/Guides/Internet/Evaluate.html). The UC Berkeley Library has a Web page evaluation check list (authorship, publisher, current content, etc.) to help you determine the credibility of a site.

When I do searches on an unfamiliar topic, I often seek a second opinion. I went for a second opinion on evaluating Web sites and came across additional information on "Evaluating Information Found on the Internet" (http://www.library.jhu.edu/researchhelp/general/evaluating/), which is located on the Johns Hopkins University, Sheridan Libraries Web site. The JHU site provides the following considerations to help you determine the credibility of the information.

- Authorship.
- Publishing body.
- Point of view or bias.
- Referral to other sources.
- Verifiability.
- Currency.
- How to distinguish propaganda, misinformation, and disinformation.
- The mechanics of determining authorship, publishing body, and currency on the Internet.

Exchanging information on the Internet via discussion lists

To exchange information and discussions via the Internet, you can communicate in a wide variety of formats. In addition to discussion lists, other methods are chat, instant messages, newsgroup, forum, conferencing, video, and IP telephony.

There are two kinds of discussion lists. One format is for announcements only, and is conducted one-way. One person (i.e., the manager or owner) sends a message to the server and it goes to all subscribers. In contrast, there is a two-way discussion list like the Seafood HACCP Discussion List (through the University of California Sea Grant Extension Program) where every subscriber receives and has a chance to send a message that is distributed to everyone on the list.

There are some disadvantages and advantages to discussion lists. Sometimes an interesting topic is posted, but no responses are publicly posted. Some subscribers may be too timid to respond publicly. Yet, there is likely to be communications occurring behind the scenes. (One subscriber posted a message on the Seafood HACCP Discussion List and received 10 personal replies.) Not all the topics are of interest to subscribers. The archive is not categorized by topic, so it is a little hard to find information from the past. Some subscribers may go off on tangents. The FDA seafood list has over 1,500 species and there are many different kinds of processes, packages, and end uses—so there are many unique processing circumstances, which may not all be of interest to your own situation as a smoker. As a result, you may find that you are receiving messages at times that are of no interest to you.

Discussion list disadvantages:

- Information may be incomplete; some posts are ignored or answered personally instead of shared publicly.
- Not all topics are of interest to the subscriber.
- On large lists, error messages (i.e., server down) may return to the person posting a message.
- Potential for receiving frequent email messages.
- Subscribers may go off on tangents.
- Topics, if archived, may be chronological and challenging to retrieve information.

Some of the advantages of discussion lists are that they are fast, accessible, and inexpensive; they allow people with common interests to discuss topics via email to a large group; they provide assistance and resources for risk analysis activities; and they offer rapid opportunity for the exchange of information. The Seafood HACCP Discussion List offers free assistance worldwide to the seafood community.

Discussion list advantages:

- Accessible and inexpensive.
- Allows people with common interests to discuss topics via email to a large group.
- Assistance and resources.
- Real time (rapid) communications and exchange of information.

I manage two kinds of discussion lists. One is "announcements only" with over 200 subscribers on ca-seafood@ucdavis.edu. Most of the information that I share is of interest to California processors. In the past I included information on a judge's ruling on mercury labeling in canned tuna that was pursued by the California Attorney General. When there are workshops, reports, new regulations, etc., of interest to the California industry, they receive notification via the announcements only list.

The other kind of listserv that I'm in charge of is the Seafood HACCP Discussion List, which has over 1,000 subscribers worldwide consisting of processors, inspectors, importers, retailers, fishermen, trade associations, educators, and others. Over 55 countries are represented. The Seafood HACCP Discussion List covers primarily seafood processing, HACCP, and regulatory topics. The Seafood HACCP Discussion List was created in 1995 when the seafood HACCP regulation, which would impact the seafood industry in a new and significant manner, was soon to be announced. The list was designed as an open Internet forum for communicating processing, HACCP, safety issues, conferences, and career opportunities to the global seafood community. The intent was to exchange information among processors, inspectors, Sea Grant specialists, etc., in order to successfully adopt and implement the U.S. Food and Drug Administration's new HACCP regulation.

When you are subscribed to the Seafood HACCP Discussion List, you may get several email messages weekly or you may get several messages in a day. The discussion list software has an option that can group several daily messages and thus consolidate all the messages into one bundled message per day compiled in the digest (http://seafood.ucdavis.edu/listserv/listinfo.htm).

Occasionally the Seafood HACCP Discussion List covers smoked seafood discussions. Following are smoked seafood topics on the Seafood HACCP Discussion List between 2000 and 2007:

- Effect of smoke on *Listeria monocytogenes* (2000).
- Use of lactates in smoked products (2000).
- Guide to making safe smoked fish (2000).
- Cold smoked critical control points (2000).
- Smoked salmon, pH and a_w (2000).
- Nitrite in cold smoked salmon (2006).
- HACCP question on commingling hot and cold smoked products in the cooling room (2007).

You can go to the chronological archives on the Internet and view the information without having to subscribe (http://listproc.ucdavis.edu/archives/seafood/). The archive is not categorized by topic, so it is a little hard to find the information from the past.

To find a topic that was posted on the Seafood HACCP Discussion List, you can also visit the UC Davis Web site and post your key search words in the search box that is located in the upper right corner, at http://www.ucdavis.edu. The Google search engine on the UC Davis site will provide links to all the topics discussed on the Seafood HACCP Discussion List related to the search term(s) that you enter in the search box.

There are other kinds of food related discussion lists available on the Internet. If you go to http://seafood.ucdavis.edu/pubs/onlinenews.htm, you can link, subscribe, or get more information on these other lists.

Key Web sites worth bookmarking

I also own and manage the Seafood Network Information Center (SeafoodNIC) Web site (http://seafood.ucdavis.edu). SeafoodNIC averages about 1,000 visitors daily from all parts of the world. Many of the topics are of interest to the seafood industry. An easy way to find out about HACCP courses is to go to this Web site and click "coming events" on the left panel.

There are numerous smoking topics at SeafoodNIC (http://seafod.ucdavis.edu/pubs/bysmoking.htm) including the "*Listeria monocytogenes* Control Manual, Smoked Seafood Working Group of the National Fisheries Institute and National Food Processors Association (2002)." This site also has his three smoking PowerPoint presentations from Dr. Martin Wiedmann's team at Cornell University (*Listeria* Controls in Finished Product [Higher Risk] Areas, Cross Contamination; *Listeria* Controls for Smoked Fish; and Plant Cleaning and Sanitation to Control.

The Seafood HACCP Alliance wanted a compendium as a companion to the HACCP training curriculum and the U.S. Food and Drug Administration's F*ish and Fisheries Products Hazards and Controls Guidance* manuals. The *Compendium of Fish and Fishery Product Processes, Hazards, and Controls* (http://seafood.ucdavis.edu/haccp/compendium/compend.htm), with 28 chapters, is available only through SeafoodNIC. It is divided by seafood process and controls and the three hazards (biological, chemical, and physical). Each chapter describes the potential hazards and provides information on how you can implement controls. The compendium also provides other resources to help you develop your HACCP plan.

Chapter 7 on "Smoked Fish and Fisheries Products" would be of interest to most of the attendees at the International Smoked Seafood Conference.

SeafoodNIC also has information devoted to seafood HACCP (http://seafood.ucdavis.edu/haccp/ucd.htm).

If you are looking for information on regulations or issues addressing smoked fish or other kinds of seafood, check out http://USA.gov. The site has resources for federal, state, local, and tribal governments. You would type your question or key words in the search box and then click "search."

The Food and Drug Administration has a useful site (http://www.fda.gov/oc/industry) with links to all of its regulatory issues. The site includes the Code of Federal Regulations, guidance documents, analytical methods, personnel directory, and much more.

As processors, smokers need to follow the HACCP regulation. If you are developing or re-evaluating your HACCP plan, which should be done annually, you may want to verify the critical control points in your own operation. Reviewing the online warning letters will help you understand some of the common violations that smoked seafood processors need to improve. What are these violations and what can you do to avoid getting written up by the seafood inspector? By conducting a search with http://USA.gov and using key words (FDA warning letter smoked fish), the search yielded 448 results documenting the smoked fish violations. The violation is noted along with a citation of the regulation. Many violations among smoked fish operations fall under failures in the following areas: GMPs (good manufacturing practices), brining, smoking, and finished product storage critical control points.

The FDA's "Seafood Information and Resources" site (http://www.cfsan.fda.gov/seafood1.html) houses *Fish & Fisheries Products Hazards & Controls Guidance*: 3rd Edition, June 2001. This is a key resource that is recognized by the FDA as scientific documentation for processors in developing and maintaining their HACCP plan. A revised version is expected in 2008, and should be available at this site and also via SeafoodNIC.

For those interested in sourcing for information for product development projects, one database that could be useful is the National Agricultural Library (http://agricola.nal.usda.gov/), which has links to books, journal articles, book chapters, short reports, reprints, and others. Use different combinations of search terms to broaden your search such as "smoked fish," "smoking fish," "smoked seafood," "smoked salmon," etc. While the NAL does not loan out the documents to you personally, you can obtain these materials via inter-library loan. Check with your local library for details on inter-library loans.

The FAO sponsors a Web site called OneFish (http://www.OneFish.org). There are at least 11 documents related to smoked seafood with substantial discussions.

The International Smoked Seafood Conference was cosponsored by Sea Grant (http://www.seagrant.noaa.gov/colleges/), a federally funded program with 30 programs on the West Coast, Gulf Coast, East Coast, the Great Lakes, Puerto Rico, and Guam. The National Sea Grant Library (http://nsgd.gso.uri.edu/) is the archive for Sea Grant–funded documents. This collection covers a wide variety of subjects, such as oceanography, marine education, aquaculture, fisheries, aquatic nuisance species, coastal hazards, seafood safety, limnology, coastal zone management, marine recreation, and law. The library maintains an online, 40,000-record, searchable database containing citations and abstracts of Sea Grant publications and, in many cases, provides access to full-text electronic copies. I keyed in smoking fish and smoked fish and came up with 33 titles. Many of the documents are in PDF format. You can also borrow materials from the library.

Many of the speakers on this Smoked Conference program are Sea Grant specialists and advisors: Alaska Sea Grant (Donald Kramer, Liz Brown, and Chuck Crapo), California Sea Grant (Pamela Tom), Delaware Sea Grant (Doris Hicks), New York Sea Grant (Ken Gall), and Virginia Sea Grant (Michael Jahncke). Other Sea Grant programs with seafood technology programs include Oregon, Florida, Georgia, Louisiana, Maryland, North Carolina, Rhode Island, and Texas.

If you don't have access to the Internet or cannot find information on the Internet, I encourage you to contact the Sea Grant seafood specialist or advisor in your state so that you can get help with your seafood technology information needs.

Conclusion

In conclusion, the Internet is a wonderful and powerful tool for sourcing scientific and regulatory information instantaneously. Having access to the Internet at work is like having the library at your fingertips. I gave an overview on search engines, advice on how to evaluate the credibility of the site that you visit, information on Internet discussion lists, and a number of reliable resources for accessing smoked seafood information.

Planning, Constructing, and Managing a Community Freezing and Cold Storage Facility: A Case Study

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In August 2006, the Petersburg Economic Development Council completed construction of the Petersburg Community Cold Storage Facility (PCCS) after several years of planning and fund-raising. As the facility enters its second season of operation, changes continue to be made to its physical structure and operational procedures. This brief paper describes the facility and how its construction was funded, provides the estimated operating expenses, and presents the challenges encountered in a community-run entity such as this.

As built, the PCCS has a total storage capacity of 450,000 pounds. It also holds three custom-built blast freezers capable of freezing 75,000 pounds per day, as well as a smaller custom-built plate freezer capable of freezing 5.000 pounds per day.

It also contains a small ice machine, which can produce about 500 pounds per hour. The entire facility uses a 6,000 square foot footprint, and includes a mezzanine. It is located on property leased from the City of Petersburg adjacent to the South Harbor.

Funding for construction of the project came from two sources. The first was a $1.3 million grant from the Alaska Department of Commerce, Community, and Economic Development (DCCED), granted in 2003. In October of that year, the citizens of Petersburg voted to allow the use of $500,000 from the Petersburg Economic Development Fund for the project. An additional award of $460,000 was granted by DCCED in 2004. Given this limited budget, the decision was made to have the facility built under a "design/build" contract, which we estimate saved over $300,000.

Operating expenses are expected to be $225,000 per year. This includes electricity at $0.1025 per kwh and salary for one full-time plant manager and two seasonal employees. Income will come from blast freezing and cold storage fees, as well as ice sales. Freezing rates will be $0.07 per pound for freezing and glazing, and storage fees are $30 per standard tote per month. Depending on the product stored, this works out to about $0.03 per pound per month. Ice is sold at market rates, generally $80-$100 per ton. Conservative early projections have the facility operating in the red for the first four years, but the facility can break even if it freezes 1 million pounds of product and remains 80% full all year.

The PCCS is owned by the Petersburg Economic Development Council (PEDC), an independent nonprofit funded by the City of Petersburg. PEDC is a nine-person volunteer board that oversees one full-time economic development director. After funding for the facility was secured, the PEDC appointed a cold storage oversight committee that oversaw construction of the facility, oversees the plant manager, and sets cold storage policy with approval from the PEDC board.

In addition to the difficulties of securing funding and overseeing construction, we anticipate future challenges with the operation of a community-based facility such as PCCS. The largest of these will be balancing the needs of larger users, whose product is needed to keep the facility open, with those of the smaller, more community-based users. The PEDC and oversight committee are committed to ensuring that small users have access to the facility, but also need to be fiscally responsible. Having carefully considered but flexible policies should help address this challenge.

Additional challenges may include the cold storage area being too small for demand as more users are attracted to the facility. The location of the facility is not ideal, being on industrial-zoned land immediately adjacent to residential areas. The facility is also close to, but not directly on, the waterfront. Finally, cash flow through the winter may be problematic.

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RFID: How It Will Transform Packaging, Distribution, and Handling of Alaska Seafood

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This paper is about Radio Frequency Identification (RFID). RFID is used for automatic identification and data collection on small tags.

There is a famous quote attributed to the hockey player Wayne Gretzky—when somebody asked what makes him such a good hockey player, he replied "I don't go to where the puck is, I go to where the puck is going to be." RFID is one possible element of the future seafood industry, and I hope it might become a useful tool for you.

RFID is used for automatic identification of a product, and it's used for data collection. It's similar to a bar code, except that it stores data, and it uses radio frequencies instead of light waves so it doesn't have to be in line-of-sight. The tag consists of an RFID antenna, and a tiny chip. The chip does not contain a great deal of data, but enough data to make it useful to us.

Why we need RFID

Visibility is critical to effective logistics support. We need RFID because there are all kinds of items in our supply chains. Whether it's motor oil, clothing, or fishing lures, things get lost and we want to know what they are, where they are, and what condition they're in.

Who is affected by RFID?

One of the biggest proponents of RFID is the U.S. Department of Defense. They deal with so much materiel that they have trouble keeping track of it all. And it comes from many vendors. So DoD has mandated RFID on virtually everything they buy. It can be used on tanks, down to boxes of bullets.

RFID is already being used in other parts of the supply chain. Wal-Mart is also a big user of RFID. So if you are a supplier to DoD or Wal-Mart, RFID policy impacts you. Besides DoD and Wal-Mart, many other companies are already implementing RFID, at the behest of their internal management, or at the behest of their customers: Target, CVS, Farm Fresh, Firestone, Michelin, Boeing, Gillette, P&G, TESCO, Georgia-Pacific, Unilever, Kimberly-Clark, Johnson & Johnson, General Mills, Marks & Spencer, PEPSICO, and others. Everybody will become affected by RFID in one way or another.

Wal-Mart is very serious about RFID. They've used it on pallets, and they've used it on cases. It's visible at the receiving and shipping doors. They're ordering about 10 billion tags per year.

Components of the RFID system

A basic RFID system consists of the following:

- Computer: storage and processing of data.
- Middleware: software required to control large amounts of constantly streaming data.
- Reader: device that interacts with tag data and computer outputs, and transfers data to computer.
- Reader antenna: emits RF for tag activation; its design is important to system effectiveness.
- Tag: contains data to identify item of interest, and communicates with reader.

Tag types: active vs. passive

There are two basic kinds of RFID tags. An *active tag* is battery powered, and is always sending out a signal. If an antenna is nearby, the antenna can pick up the signal. Active tags are predominantly used in transportation systems. It has an internal power cell.

Active tags

- Standard: none, mainly manufacturers' proprietary systems/protocols (transmits RF energy in the 400MHz, 900MHz, and 2.45GHz ranges).
- Range: generally 300 feet or less (battery replacement).
- Used predominantly in transportation systems (rail, toll systems, trucking, container).
- Characteristics: tag with internal power cell mounted to item or container/pallet/box, interrogator queries tags, uploads/downloads data. Do not transmit all of the time. Data capacity varies.

A *passive tag* just sits there until it is "pinged" by RF signal, and it answers back "Yes I am a case of motor oil, but I'm not just any case of motor oil, I am the 375,123rd case of motor oil produced by Pennzoil this year." RFID is not just product identity, it's individual package identity.

Passive tags

- Standard: none, mainly manufacturers' proprietary systems/protocols (uses back-scatter technology).
- Range: typically measured in "inches," industry working toward "meters" (dependent system layout, interference, etc.).
- Used predominantly in retail systems and transportation systems.
- Characteristics: small tag loaded with license plate data, typically mounted to end item, reader captures data as item moves through choke point (door, pathway, frame, etc.). Could have a battery. Data capacities are limited.

Tag types: read vs. read/write

There are other ways to subdivide RFID tags—"read-only" vs. "read-write." Information that can be on an RFID device is programmed at manufacture, compared to a user-programmable which picks up information as it moves along. That's especially useful in a value-added or complex manufactured products where the product is modified as it moves along.

For *read only* tags, information can only be read from an RFID device; it is programmed at manufacture. Read only tags are user programmable. They are "WORM"—write once read many. The RFID device can be initialized outside of the RFID manufacturer's facility after manufacture.

For *read/write* tags, information can be read from or written to an RFID transponder during the time it is presented to a reader/writer. These typically have an asymmetric read and write operating range.

Electronic product code—EPC

The electronic product code is a 96-bit code created by the Auto-ID Center. Founded in 1999, the Auto-ID Center is a unique partnership among almost 100 global companies and five of the world's leading research universities. Together they are creating the standards and assembling the building blocks needed to create an "Internet of things."

RFID standards have been developed for a couple of decades. They rely on an electronic product code (EPC); it is similar to a UPC, but there's a lot more capability in the EPC. The electronic product code will one day replace barcodes. The EPC has digits to identify the manufacturer, product category, and individual item. It is backed by the United Code Council and EAN International, the two main bodies that oversee barcode standards.

EPC 96-bit code

- Version: identifies format of EPC. (Future tags may contain more than 96 bits.)
- Domain manager: manufacturer of product.
- Class code: stock keeping unit (SKU) or UPC code; i.e., 12 oz. cherry soda.
- Serial number for item; i.e., exactly which 12 oz. can of cherry soda.

Bar code vs. RFID

Some comparisons between bar codes and RFID

- Bar codes are line of sight, RFID can read through many materials.
- Bar codes have a limited read range (it has to be very close), RFID has a much longer read range.
- Bar codes are read-only while RFID has read-write capability.
- Bar codes have a limited amount of data, but RFID can hold a lot more data.
- Bar codes identify the type of product, but RFID identifies a specific item.

Identifying the specific item is especially useful in "retro logistics," or what we call "reverse logistics." In the event of a product recall, you want to know which particular product caused a problem or made somebody sick.

The business process for RFID is that there is a tag, a reader, and a computer network that this all responds to. The tag on the box or product moves from the manufacturer, to the distribution center, through the distribution chain, to the store, and then goes out of the store. When it is removed from the shelf, the sensors in the store say "We have sold X many units, this many cases of coke—order more!"

The promise of RFID

The promise of RFID is that its extremely low cost, and extremely high performance. It offers some tremendous advantages in security and visibility to the manufacturer and to the vendor, and accuracy from the point of manufacture to the retail shelf. Every package contains a unique serial number, so you know exactly which package you're dealing with. The current focus is on pallet and cases, not individual items—yet.

RFID is part of a solution. It's not just about tags, it is about a system. It's part of the vendor business process, so if you're a vendor, you have to meet those standards. If you are a vendor for Wal-Mart or Target, DoD will accept those standards.

It's a "copper bullet" not a "silver bullet." It's not a magic solution, but it's a useful solution, and it's going to become more useful as time goes by. RFID is not an IT or supply chain management solution; it is both.

By the way, if anybody's looking for career opportunities, we predict that this will be a growth industry, because it will need people to deal with all of this technology and this massive volume of data. There is an RFID personnel shortage today.

Warehouse management

RFID is useful at several points in the supply chain. It's useful in warehouse management, where you can track your lift trucks, or you can track individual seafood products. In parcel logistics, as you are moving things around, as you're shipping things from point A to point B.

With RFID you can

- Uniquely identify, collect, sort, and track more efficiently.
- Track hard data on pallets, containers, forklift trucks, equipment, and man-hours.
- Collect data in rugged environments where barcodes can't.
- Enhance productivity and reduce costs.

Utilizing RFID in manufacturing

Automatic product tracking can be accomplished through production, shipping, and after-sales service. During manufacturing, a work item with a tag attached can be tracked in the production process. This is especially important in plants that produce automobiles or military tanks, which are very complicated, multistep processes. An RFID read-write tag can tell you what's been done to product X, and if it is ready for shipment. In addition, quality control status can be tracked manufacturing.

Retail supply chain

Of course, RFID is useful in the retail supply chain, at the distribution center, and even in the retail store, and it can be used to trace returns.

RFID can be used to

- Program "contents" data at manufacturing source.
- Program "destination" data at distribution center.
- Identify whole carton contents, while the carton is closed.
- Trace returns.

Electronic article surveillance (EAS)

RFID is part of an electronic article surveillance type of thinking. One possible vision of the future is that you go to Wal-Mart or Fred Meyer, fill up your shopping cart with anything you want, and walk out of the store. As you leave the store, the portal has an antenna in it. The antenna scans all the items in your shopping cart, it scans the credit card in your wallet, and sends you a bill, which you can pay online.

New RFID technology provides significant features:

- Able to write SKU number into transponder.
- Automatic inventory with a hand-held reader.
- Anti-collision mandatory for this feature.
- Cash registers can automatically ring up merchandise.

Why RFID now?

Right now is a good time to be thinking about RFID—maybe not yet for small manufacturers, but you want to capitalize on some of the emerging supply chain technologies, especially if your customers are going to demand it. If you can influence the cost and direction of the technology, and make it useful to the seafood industry, as opposed to the big beef industry, that's going to be another advantage.

The timing is right:

- To capitalize on emerging supply chain technologies.
- To influence direction and cost of the technology.
- To make the standards work for us.
- Lay the foundation for future supply chain improvements today.

We're all familiar with how the USDA COOL Rule (country-of-origin labeling) was imposed on the seafood industry, but it has not yet been imposed on the beef industry. The seafood industry is vulnerable, in terms of being a test bed or guinea pig, so we hope to make the standards work for us.

RFID is part of a solution that includes people, process, and technology. What bar codes did to manual price tags, we think RFID will do to bar codes. But there will be some changes that we can't foresee yet.

In the basic seafood supply chain, RFID can be applied in any number of places:

A tag on a salmon, for example a Copper River salmon.

- On products leaving a processing plant.
- For brokers and traders.
- For imports, exports, and re-imports.
- Especially for "no processing," going straight to a customer.

With RFID, I can tell that the seafood that I shipped from Kenai wound up in New York in fine condition. And I'll tell you more about what that can mean to you.

You can use RFID in the seafood industry to keep track of various things.

What

- Species, product type, origin
- Package identity

Where

- Location

How

- Temperature

- Condition

Who
- Harvester
- Processor
- Carrier
- Distributor
- Customer

RFID combination technology

For those of you who have heard about RFID, I want to make a clear distinction between RFID and other types of technologies related to seafood safety and quality, such as time-temperature indicators (TTIs). For example, some people have told me that they can keep track of the quality of their products simply by tossing an RFID chip into the box, and that will tell them about time and temperature.

No, it won't. Time-and-temperature is time-and-temperature, and RFID is RFID. They are not the same. My point here is those technologies, though separate, can be linked. In the future there will be RFID tags and systems where you can sit at your desk and read how your product on its way from Omaha to St. Louis is doing, what time it got shipped, its temperature, what it's freshness condition is, and if it's been contaminated or tampered with in any way.

These combination technologies are not evolved yet, but they are evolving now, and like I said, "this is where the puck is going to be."

Anything that can be turned into an electronic signal can be put onto an RFID tag and transmitted to the person who wants to read it, who is probably you. For example, if the door on your container has been opened, there is a sensor that can tell that to the RFID chip, and the RFID chip can tell that to you.

Current technical limitations

We still have some technical limitations. It's a complex type of system, and there are some security issues that we have to deal with. When the tag leaves the store, and the great RFID computer sends you a bill, all the computer knows is that the tag left the store, it can't prove that the actual purchased item left the store. So somebody could take the tag and put it in your shopping cart and you don't even know it, and then you get charged for something you didn't actually buy. There are security issues, and there are hacker issues in the databases of RFID systems.

Some of the problems about the tags not working well in cold temperatures have been solved or are being solved right now. But if the tags are close to steel, which interferes with the signal, and this issue of battery life in the active tags, those are still evolving technologies. But the batteries in our cell phones have become a lot smaller over the years, and I anticipate the batteries in RFID tags will become even smaller than they are now. These problems will disappear.

What do you want to manage with RFID?

- Do you want to manage the manufacturing process in the plant?
- Do you want to manage your warehouse and inventory after the products are processed?
- Do you want to manage your parcels and shipments while they're in transit in the supply chain?
- And/or, do you want to know what's in the retail store someplace?

Here's the thing about Wal-Mart and some of these other retailers: they say that the vendors must manage their own inventory in the Wal-Mart stores. So the vendors have to figure out when Wal-Mart is getting ready to run out of the vendor's product, because Wal-Mart doesn't want to be bothered with taking inventory.

And RFID is an ideal solution for that problem. You can sit in your office in Seattle or Anchorage and say "Oh yes, the Wal-Mart store in Peoria needs a hundred more units of such-and-such," because your RFID tags tell you that the previous items have been sold.

Some questions on collecting, analyzing, managing, and reporting data:

- What do you want to know?
- Who wants to know it?
- When do you want to know it?
- How soon do you need to know it?
- When to apply the RFID tag to your processed product?
- Do you want to add data as the product moves in the seafood supply chain and becomes further processed?
- How to use it in product combinations like crab-stuffed flounder?
- Who's going to pay for it, and who's going to do the work?

All of these questions influence your choice of particular RFID technologies. For example, in going from an H&G salmon to a fillet portion and the crab-stuffed sole—do you want to apply a new RFID tag after that further processing?

Seafood at Wal-Mart

RFID is mandatory for Wal-Mart's top 100 suppliers, and Wal-Mart will add seafood to their RFID-monitored supply chain, and they will add "sustainable" seafood, as you know, as it is certified by the Marine Stewardship Council (MSC). So somebody's got to track that chain of custody, and RFID is a possible solution to that.

You have three strategic options

1. You can just slap-and-ship: your customer gives you their RFID tags, and you put them on your products.

2. You can start with a supply chain partner who already has RFID, puts the tag on for you, and reads the tag for you. The RFID chain starts with your partner (customer).
3. Or you can do-it-yourself, as outlined in this article.

Approach to RFID

You can learn from the seafood industry innovators, such as Beaver Street Fisheries and Fisk Seafoods, who are very active in RFID. You can also become an early adopter—establish a pilot project. In the introduction of any new technology, there are some innovators, early adopters, early majority, late majority, and laggards. You have to figure out when it will become worth it for your company.

You can incorporate RFID as a new tracking tool in individual cartons, you can hook it to the tail or gill cover of your fish, or just drop it inside an individual carton and/or the master box (especially for tracking temperature), or it can be attached to the inside or outside of a tote shipping container. Another option is a customized tag developed for a specific application.

Precautions

For startup, we recommend you do a pilot run, and see if it works for you—see if the technology is as friendly to you as you hope it will be. You want to make sure that you have selected the correct type of labels or tag, location of label or tag, and type and functionality of systems hardware and software. Work with integrators to multiple solution options.

You want to make sure that the location and the environment for the labels and the tags are conducive for RFID. Cold temperatures, being next to steel, and non-skid floors all interfere with RFID. You also want to maintain quick access to trouble-shooters and consultants.

Next step in adopting RFID

You'll want to do an internal environment scan to make sure that your company has the best solution—whether you apply the tags at a station with minimal contact with water, how the boxes are transported, and where they're stored. They must be free of metal barriers.

Internal environmental scan

- Select stations for application of tags to minimize incremental time and motion.
- Apply Tags at stations where there is minimal contact with water.
- Store and transport tagged carton/boxes in areas that allow the readers to function free of metal barriers.

Quality assurance plan

Developing a quality assurance plan for RFID can be easily integrated as one or two steps into your standard operating procedures. And that's something you can help your customer with, especially if both of you want to see the RFID data.

- Develop a quality control plan.
- Establish an action plan for rejects.
- Recommend a quality plan for the customer.

RFID costs and benefits

Costs are dependent on selecting the type of overall RFID system for an application. The system is hardware, software, and training. RFID costs are low and they're getting lower. They're becoming more affordable every day, just like cell phones and faxes.

Costs depend on

- Type of labels or tags
- Type of reader
- Type of network, server, stand-alone PC, etc.
- Costs/expenses associated with integration and configuration of the overall system.
- Other (communication systems, etc.).

The benefits of RFID are labor savings, inventory system accuracy, and automatic data communication to many interested parties.

Evaluating RFID costs/expenses

- Assess the incremental direct material cost (labels/tags).
- Establish additional direct labor cost (if any).
- Determine if there will be added cost for handling, storage, and transportation.
- Evaluate incremental cost (if any) for adding quality control steps.
- Compute costs relating to incorporating the overall RFID system (hardware, software, and integration).

Look for cost/expense savings ideas to implement RFID; you may want to use a consultant for this. Feel free to contact me or my co-author Mike Ronchetti—we're in Anchorage. Conduct a cost/benefit analysis. Based on that, make a GO/NO-GO decision. If GO, develop and implement an action plan (start with a pilot-run).

RFID and seafood—summary

- RFID is in use in many supply chains, such as DoD and Wal-Mart.
- Its use is increasing everywhere.
- RFID is a tool.
- It might be part of a solution.
- Its application to seafood, even though new, is growing.
- RFID technology and standards are evolving.

- The systems to use the technology are evolving.
- Seafood producers have many options.

RFID Web sites of interest

http://www.rfidcomplete.com
http://www.cbpp.uaa.alaska.edu
http://www.alientechnology.com
http://www.autoidlabs.org
http://www.acq.osd.mil/log/rfid/index.html
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http://www.ncseagrant.org/files/seafood_traceability.pdf

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USDA Rural Development: Value Added Producer Grant (VAPG) Program

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Value Added Producer Grants provide cost-share funding to help agricultural producers enter new markets with value-added products. USDA Rural Development, through its Rural Business and Cooperative Service, provides for a nationally competitive grant program to assist agricultural producers to expand into emerging markets. This effort is to increase on-farm income and rural economic development. The purpose of the VAPG is to help eligible applicants develop business plans for viable marketing opportunities, and develop strategies to create marketing opportunities in emerging markets.

Value-added is the incremental value that is realized by the producer from an agricultural commodity or product as the result of a change in physical state, e.g., smoked product; differentiated production of marketing, e.g., branding of a product; or product segregation, e.g., GMO vs. non-GM corn.

Eligible applicants

To be eligible, the applicant must be in one of the following groups.

- Agricultural producer (including fishers, shellfish farmers, and loggers).
- Independent producer.
- Farmer or rancher cooperative.
- Agricultural producer group.
- Majority-controlled, producer-based business ventures.

Agricultural Producers: Individuals or entities directly engaged in the production of agricultural products that obtain at least 50% or greater of their gross income from their agriculture business.

Independent Producers: Agricultural Producers, individuals or entities (including for-profit and not-for-profit corporations, LLCs, partnerships, or LLPs) where the entities are solely owned or controlled by Agricultural Producers who own a majority ownership interest in the agricultural product that is produced. May also be a steering committee of independent producers in the process of organizing as an association to operate a value-added venture.

Agricultural Producer Group: An organization that represents Independent Producers, whose mission includes working on behalf of Independent Producers, and the majority of whose membership and board of directors is composed of Independent Producers.

Farmer or Rancher Cooperative: A 100% farmer- or rancher-owned, controlled, and incorporated cooperative from which benefits are derived and distributed equitably on the basis of use by each of the owners.

Majority-Controlled, Producer-Based Business Venture: A venture where more than 50% of the ownership and control is help by Independent Producers, or held by partnerships, LLCs, LLPs, corporations, or cooperatives that are themselves 100% owned and controlled by Independent Producers.

Eligible activities

Grant funds can be used for planning and working capital. Planning is a defined program of economic activities to determine the viability of a potential value-added venture including feasibility studies, marketing plans, business plans, and legal evaluations. Working capital funds are used to operate the venture and pay the normal expenses associated with the operation of that venture.

Planning activities

- Conduct a feasibility study.
- Develop a business plan.
- Develop a marketing strategy.
- Conduct a feasibility study for renewable energy activities.

Working capital

- Pay payroll.
- Pay utilities.
- Pay normal expenses of the value-added venture.

Ineligible activities

Funds cannot be used to purchase long-term assets. Grant and matching funds cannot be used for purposes outlined in the notice of solicitation of applications (NOSA). A partial list of ineligible grant uses includes the following.

1. Plan, repair, rehabilitate, acquire, or construct a building or facility (including a processing facility).
2. Purchase, rent, or install fixed equipment including mobile and other processing equipment.
3. Purchase, repair, or maintain vehicles.
4. Purchase long-term assets.
5. Pay for the preparation of the grant application.
6. Pay expenses not directly related to the funded venture.
8. Fund political activities or lobbying activities.
9. Pay costs incurred prior to receiving a grant.
10. Fund any activities prohibited by 7 CFR parts 3015 and 3019.
11. Fund any agricultural or engineering design work for a specific physical facility.
12. Fund any expenses related to the production of any commodity or product to which value will be added.

Matching funds

This grant program has a *matching funds requirement*. Applicants must provide matching funds at least equal to the grant. Other federal grants cannot be used as matching funds. At a minimum, grant recipients must match a dollar for each dollar of grant funds. Grant funds and matching funds must be spent proportionately during the time frame stated in the grant application.

Applicants must certify that matching funds will be available at the time grant funds are received. Matching funds have the same use restrictions and eligibilities as the grant funds. Matching funds must be spent at the rate equal to or greater than the rate of grant funds requested.

Matching funds include

- Cash, including salaries, plant wages, utilities, and other direct cash payments, verified by a bank statement submitted with the application.
- Confirmed funding commitments for non-federal sources.
- In-kind contributions that conform to the provisions of 7 CFR 3015.50 and 7 CFR 3019.23. Examples are donated professional and technical services, labor, supplies and office equipment, and office space.

Grant terms, FY 2006

- No minimum.
- $300,000 maximum for working capital.
- $100,000 maximum for planning grants.
- Nationwide: $1.5 million is set aside for applicants requesting $25,000 or less.
- Grant funding: up to 50% of total eligible costs.
- Grant period: 12 months.

Selection factors

Criteria for Planning Grant applications

- Nature of the proposed venture
- Qualifications of those doing work
- Project leadership
- Commitment and support
- Work plan/budget
- Amount requested
- Project cost per producer
- Business size
- Number of grants
- Presidential initiative on bio-energy
- Administrator points

Criteria for working capital applications

- Business viability
- Customer base/increased returns
- Commitment and support
- Management team/work force
- Work plan/budget
- Amount Requested
- Project cost per producer
- Business size
- Number of grants
- Presidential initiative on bio-energy
- Administrator points

How to apply

Applications are selected on a nationwide, competitive basis. NOSA, forms, etc., are available from the VAPG Web page, http://www.rurdev.usda.gov/rbs/coops/vadg.htm.

Applications include the following.

- Title page.
- Table of contents.
- Executive summary.
- Eligibility discussion of applicant, product, and project.
- Proposal narrative: no more than 35 pages as per specifications.
- Verification of matching funds: bank statement, in-kind letter verifying types and availability of donations.
- Certification of matching funds: signed page certifying the expenditure of matching at or greater than the requested grant funds.

When to apply

Web resources for information regarding these notices are http://www.rurdev.usda.gov/ and http://www.usda.gov.

How much money is available?

Funding in FY 2005 was over $14 million, and FY 2006 was $21 million. For FY 2007, the amount of funds estimated to be appropriated for this program by Congress is just over $19 million.

What Is Alaska's track record?

Alaska has had four awards since 2001, totaling over $450,000, but has had no awards since 2004. In FY 2006, 186 grants were awarded varying from a $3,000 award in Michigan to several $300,000 awards nationwide.

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Resources

- USDA Rural Development Web site for Alaska, http://www.rurdev.usda.gov/ak.
- USDA Rural Business and Cooperative, http://www.rurdev.usda.gov/rbs/coops/vadg.htm.
- Agriculture Marketing Resource Center, http://www.agmrc.org.
- U.S. Forest Service Proceedings: Linking Healthy Forests and Communities Through Alaska Value-Added Forest Products, http://www.fs.fed.us/pnw/pubs/gtr500.
- UAF Cooperative Extension Service, http://www.uaf.edu/ces/dir_info/cesdirectory.pdf.
- UAF Alaska Sea Grant Marine Advisory Program, http://seagrant.uaf.edu/map.

Innovations in Alaska Smoked Seafood as Showcased through the Alaska Symphony of Seafood

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Abstract

Since 1994, innovative products with Alaska salmon and seafood have been showcased before professional judges and the public in the annual Alaska Symphony of Seafood (previously Alaska Symphony of Salmon 1994-1999). Smoked products ranging from traditional hot and cold smoked varieties, to lox, kippers, dried sprinkles, and caviar have all placed well as individual products. Smoked products have also proven successful when combined with dairy products such as cheeses, butters, and chowders. Innovations in packaging have further enabled smoked fish products to be marketed to a broader range of consumers and reach local and distant markets. This paper will highlight the innovative approaches shown through the Symphony of Seafood competition and the proportion of industry smoked products versus other new products.

History

Smoked fish has been an integral part of diets for centuries, as smoking and curing provided an option for long term storage of highly nutritional fish. In Alaska, the history of smoked fish, particularly salmon and halibut, runs deep within the Alaska Native culture as it insured a source of protein through the winter months and between fisheries. As Alaska began commercial harvest of salmon in the 1880s, salting was the dominant method of preserving. By the turn of the century, canning dominated and continues to be a major processing effort to date. Smoked salmon remained a component of the industry for local consumption, as the majority of salmon was shipped as canned fresh, while a limited amount evolved to "kippered" canned salmon. With improvements in fresh handling and packaging, the availability of smoked salmon has increased significantly. This allowed for varying levels of cure with hot and cold smoked products. Today the marketplace is filled with a large variety of smoked salmon products, produced by a broad range of commercial processors, from specialty processors to large corporations.

Like Alaska salmon, Alaska halibut and sablefish have been commercially exploited since 1880 as a primary and secondary fishery, respectively. Both fish were recognized for the ability to preserve the flesh through hot smoking. This allowed the fish to enter domestic and international markets. Today, smoked halibut and sablefish are available throughout the marketplace, although produced on a significantly smaller scale than smoked salmon.

Alaska Symphony of Salmon

The Alaska Fisheries Development Foundation recognized that a contest, in which new seafood products would compete against each other, being evaluated by a panel of professional judges and the public, would both stimulate innovation in the seafood industry and promote Alaska salmon and seafood to consumers. In support of this interest, AFDF commissioned the "Alaska Symphony of Salmon" in 1994 to showcase new products available for the retail, food service, and gift/specialty markets. The choice of which category in which to enter the salmon product remained with the company. At first, smoked salmon was regularly used as an ingredient in each category. Based on the predominance of smoked salmon products entered, the gift/specialty markets category evolved to the smoked product category. The Alaska Symphony of Salmon continued through 1999.

In 2000, the innovative product categories were broadened to allow for any Alaska seafood to be entered into the categories. This allowed whitefish and shellfish products to be entered including pollock, cod, halibut, sablefish, and crab. The transition from Symphony of Salmon to Symphony of Seafood showed an interesting pattern in the products entered. Table 1 shows the variation in the use of other seafood versus salmon for the period from 1999, the last year for the Symphony of Salmon, through 2007, the most recent Symphony of Seafood. Products from species other than salmon ranged from 56% of the entries in 2001, to as low as 24% in 2007, and averaged 40% of the entries during that time period.

Smoked seafood

Table 1 also details the smoked seafood entries for the same period. Smoked products have ranged from a low of 22% in 2002 to a high of 57% in 2007. Salmon has dominated the smoked product category, with products resulting from various hot and cold smoking techniques, variation with spices that often reflect market trends based on consumer taste, and techniques in combining salmon with other ingredients to make non-traditional products, including sausages, cheeses, spreads, and chowder/chili.

Smoked fish have been a popular segment of the Symphony of Salmon and Symphony of Seafood since the beginning. During the period 1998-2003, smoked salmon products won the grand prize each year, even though the number of smoked product entries declined. These prod-

Table 1. Alaska Symphony of Seafood entrants for salmon and other seafood as smoked or non-smoked products.

Year	2007	2006	2005	2004	2003	2002	2001	2000	1999
No. of entrants	21	19	16	14	17	18	16	32	23
No. of salmon	16	11	11	8	9	11	7	20	23
No. of other seafood	5	8	5	6	8	7	9	12	0
% Salmon	76%	58%	69%	57%	53%	61%	44%	63%	100%
% Other seafood	24%	42%	31%	43%	47%	39%	56%	38%	0%
No. of non-smoked	9	12	12	9	12	14	12	20	12
No. of smoked	12	7	4	5	5	4	4	12	11
% Smoked	57%	37%	25%	36%	29%	22%	25%	38%	48%

The event was called Symphony of Salmon from 1994 to 1999, and Symphony of Seafood 2000 and following.

Figure 1. Smoked salmon provides a variety of approaches in packaging and presentation.

Figure 2. Innovative uses of smoked salmon, such as smoked salmon chili cheesecake, are extending the range of smoked salmon and seafood usage.

ucts were traditional hot-smoked king or sockeye salmon in various styles, as shown in Fig. 1. While more varieties of smoked seafood entries were available to the judges and the public, traditional hot smoked salmon products continued to dominate, demonstrating the strong, broad popularity of this product. Changes in spices, packaging, and presentation differentiated these products. In 2004, changes in packaging and processing of fresh salmon introduced a new line of products that were favored by both judges and the public. Specialty handling and marinating of fresh salmon into products such as salmon "chorizo" and marinated sockeye tenderloins became of greater interest, temporarily overshadowing the traditional smoked salmon products. In 2007, smoked fish returned to win the Grand Prize in the form of a cold smoked halibut lox. Smoked halibut was an entry previously in 1999 as a part of a Grand Prize winning lox sampler, and in 2000 as a stand alone cold smoked lox. In each case, the cold smoked approach proved successful for flesh with lower oil content. In 2007, the effort in slicing and packaging showed the value of a smoked halibut over salmon in the changing restaurant and consumer marketplace.

Innovations in packaging have extended the shelf life of smoked salmon products, which allows further secondary preparation for a high quality presentation. Whether arranged in strips or as whole filets, presentation in unique packaging (including netting) allows the product to portray the strong "wild" image that Alaska salmon products are recognized for. Innovative uses of spices and curing have opened additional markets with flavors including Caribbean, Cajun, and teriyaki. Sugars have helped to maintain traditional flavors and have allowed the term "candy" to be applied to hot smoked salmon versus the traditional cured "candy" recognized in rural Alaska. The combination of curing, spicing, smoking, handling, and packaging, and the opportunity for

Figure 3. Alaska smoked salmon sprinkles is an alternative food topping for soups and potatoes.

high quality presentation have expanded the already significant market share that Alaska smoked salmon and small specialty processors have developed.

The Symphony of Seafood and its predecessor have offered a valuable judged event and market oriented focus group encouraging innovation with smoked salmon as a primary ingredient versus an end product. Over the last decade, processors have incorporated smoked salmon in a variety of chowders and chili for retail and food service markets. Blended chilled products have been created that have included smoked salmon caviar pate (smoked flesh and roe), smoked salmon butters, various smoked salmon dips and spreads with myriad spices, smoked salmon salads, smoked salmon cheese and cheese spreads, and a smoked salmon chili cheesecake (Fig. 2.), all tailored to the retail market. Smoked salmon sauces, for both the retail and food service markets, have incorporated smoked flesh into heated alfredo and other pasta sauces. Innovation with these products has opened product categories that are well accepted as part of a specialty "niche" market.

Incorporating smoked salmon into the processed meat alternative market has found significantly more challenge. Use of smoked salmon as a pizza topping, sprinkles, and jerky have found reasonable levels of success as a sustainable market product. In 2002, smoked salmon sprinkles as shown in Fig. 3. were particularly well received as an alternative for various food toppings. Salmon jerky of various brands and flavors continues to expand the marketplace, bringing spice, heart-healthy flavor, and locale as part of the intrigue. However, outside of these success stories, incorporating smoked salmon into processed meat products, including sausages, deli meats, nuggets, and portions have met with limited success. This is guiding product development. Efforts to incorporate fresh salmon into such products have proven difficult, other than "salmon burgers."

In looking to the future with the Alaska seafood industry, smoked seafood will continue to increase in market share as advances in packaging and processing methods that result in extended shelf life are discovered. While traditional approaches are likely to remain popular in the market place and specialty processors continue to provide upscale niche products, other trends are emerging. Lightly smoking and curing appears to be an evolving trend, as seen in the retort-pouched, smoked pink salmon for both the domestic and international markets. Lightly smoked salmon salad is penetrating the fast food market, particularly as an ingredient for sandwiches. Smoked salmon will continue to be included in sauces, particularly with the increased interest in heart-healthy and diverse diets. Opportunities will continue with traditional hot and cold smoked products, such as halibut lox finding a place as accent or accompaniment for dishes, and lightly smoked sablefish finding center of the plate acceptance in upscale restaurants. Smoked roes for caviar provide an interesting potential market given the increased consumer power in Eastern Europe. Smoked shellfish may have a future opportunity to expand with Alaska's resources; this will depend on having a stable market supply that exceeds the present demand for fresh clams and oysters. In each case, entering the market with an innovative product can prove successful when independently judged and provided to a large cross section of seafood interests. The Alaska Symphony of Seafood continues to provide such an opportunity for showcasing successful smoked seafood products.

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Alaska's Ocean Bounty. A laminated 11 × 17" placemat/poster. SG-ED-31, 10 or more $5.00 each.

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Developing Cooperatives for the Alaska Seafood Industry. G. Haight, A. Crow, and H. Geier, 2007, MAB-61, 45 pp., $10.

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