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TROPICAL AND SUBTROPICAL
SEAFOOD SCIENCE AND TECHNOLOGY SOCIETY
OF THE AMERICAS



Papers & Abstracts

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November 3-6, 1996

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SEAFOOD SCIENCE AND TECHNOLOGY SOCIETY
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(previous Tropical and Subtropical Fisheries Technology Society of the Americas)

November 3-6, 1996
Clearwater Beach, FL

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The Seafood Science and Technology (SST) Society schedules their annual conference based on a three year cycle; Year 1 - Domestic meeting in the southeastern sector of the continental United States; Year 2 - International Meeting outside the continental United States; and Year 3 - Joint meeting with the Atlantic Fisheries Technological Society alternating in locations between the southeastern or northeastern sector of the continental United States.

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COOLING RATE OF SHUCKED OYSTER MEATS IN ONE-GALLON PLASTIC CONTAINERS

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INTRODUCTION

Potentially hazardous foods are those foods that are capable of supporting rapid and progressive growth of infectious or toxigenic microorganisms or the growth and toxin production of *Clostridium botulinum*. The FDA Food Code (USDHHS, 1993) requires that potentially hazardous food be held under refrigeration at 5°C (41°F) or below or at temperatures above 60°C (140°F). In general the maximum time potentially hazardous foods are permitted to be held in the hazardous temperature range is four hours.

Raw molluscan shellfish are considered as a potentially hazardous food, but the guidelines which govern their processing and storage are contained in the National Shellfish Sanitation Program, Manual of Operations, Part II (USDHHS, 1995). The time live shellfish as shellstock can remain outside refrigeration after harvest depends upon the time of the year or temperature of the harvest water and their intended use (i.e. for raw consumption or for shucking). Once shellstock are placed under temperature control, the storage area must be continuously maintained at 7.2°C (45°F) or below. After initial refrigeration, shellstock may not remain outside temperature control for more than two hours. Following shucking, the meats must be delivered to the packing room within one hour. Shucked meats must be cleaned, thoroughly drained and packed using a schedule which permits the meats to be chilled to an internal temperature of 7.2°C (45°F) within two hours of delivery to the packing room.

Appendix C, Part II of the Manual (2) provides a series of cooling curves for oyster meats packed in various size containers to assist processors in developing cooling schedules to comply with the two hour cool down time. These curves were generated more than 30 years ago with metal containers at the request of the U.S. Public Health Service by the American Can Company. The data from these curves

2

have been extrapolated to suggest it requires two hours for meats in one gallon metal containers to cool from 10°C (50°F) to 7.2°C (45°F).

Today, plastic containers are the industry standard for packing shucked meats. The types of plastics used in the manufacture of food containers have lower thermal conductivity than metals used to construct cans. This study was undertaken to produce new cooling curves for oyster meats packed in one-gallon plastic containers which the industry can use to set cooling schedules. These curves are compared with those published in Appendix C, Part II of the Manual (2). Additionally, the cooling curves for dry pack oysters and oysters packed with water are contrasted because some processors contend that dry pack oysters cool slower resulting in a poor quality product.

MATERIALS AND METHODS

Container

The one-gallon plastic container most frequently used by the Gulf oyster industry is the "tall gallon bucket." This container and snap-on lid are constructed of high density polyethylene with a wall thickness of approximately 0.041 in. (1.04 mm). Approximate dimensions are top diameter 7 in. (177.8 mm), bottom diameter of 5.5 in. (139.7 mm) and overall container height of 7.94 in. (201.6 mm). The containers used in this study were manufactured by Venture Packaging Incorporated, Moroville, OH 44847-0246, item number LT700128.

Temperature measurement

Temperatures were measured with mercury-in-glass thermometers with the bulbs positioned at the approximate geometric center of each container. Each thermometer was calibrated against a certified thermometer over the range of 4.4 to 26.7°C (40 to 80°F). The temperature in each container during cooling was also continually recorded with a thermistor linked to an M160 data logger (Omega Engineering, Stamford, CT).

Oysters

Oyster meats were purchased from a local shop on the same day they were shucked. The meats were adjusted to the desired temperature before initiating each cooling experiment as follows. The contents of each gallon container of shucked meats was distributed into four one-gallon zip-seal bags. The bags were suspended in approximately 20 liters of water at the desired temperature for one hour. The

meats were removed from the bag and drained for two minutes on a standard skimmer before placing in the test container.

Ice

Commercially-produced flake ice was obtained the day prior to the study and held overnight in a cool room at 4°C (39°F).

Experimental procedure

Oyster meats were adjusted to the desired experimental temperature, as described above, and placed in containers, two for each study. Lids were placed on each container and thermometers and thermistors were inserted through holes in each lid to the approximate geometric center of the container. The initial temperature was recorded and the containers were placed onto a two-inch thick bed of ice in an insulated box and then surrounded with wet ice. Lids were covered to a depth of about one inch with ice. The insulated box was equipped with a drain to remove water resulting from the melting of the ice. Temperature readings on the thermometers were recorded at 30-minute intervals. As needed, fresh ice was added to the chest.

After each experiment was completed, the meats were removed from the test container and adjusted to the new starting temperature.

Comparison of metal and plastic containers

Data was obtained by using plastic containers. It was compared with the data taken from the curves provided on page APC-2, Appendix C, Part II of the Manual (2). Those curves were developed by using oysters packed in (610 x 708) metal cans.

Solid vs. wet pack study

In recent years, some Gulf processors have begun packing shucked oysters by the weight of drained meats. A common packaging weight is 6 pounds, 7 pounds or 8 pounds of drained meats in a one-gallon container with water added to fill the container. An 8-pound gallon would represent a "solid" or "dry" pack. For the solid vs. wet pack study, the cooling rates of an 8-pound gallon (no water added) and a 6-pound gallon (6 pounds of oyster meats and water to fill the container) were compared.

RESULTS

Figure 1 presents the results of five separate cooling studies in which the initial temperature of the oyster meats ranged from 9.4 to 25.6°C (49 to 78°F). Data for each study represents the average of two replicates. In all trials there was an initial lag of 0.5 to 1 hour after icing before the temperature at the center of the container began to decrease. Subsequently, the temperature decreased at a rate of 2.8°C (5°F) or less per hour. As the temperature decreased in the container, the rate of temperature decrease declined. Only packs of oyster meats with an initial temperature below 10°C (50°F) were cooled to 7.2°C (45°F) at the geometric center within two hours of icing.

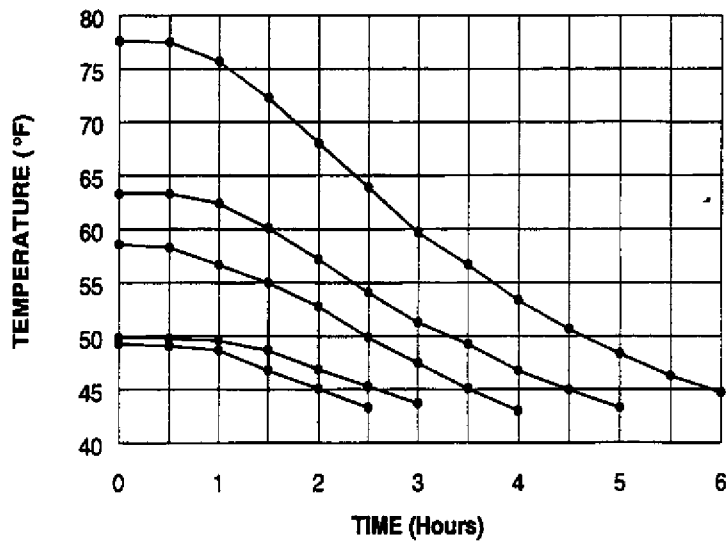


Figure 1. Cooling curves for oyster meats in the geometric center of one-gallon containers packed in ice

A comparison of cooling curves for meats in one-gallon plastic and metal containers is shown in Figure 2. At the two starting temperatures shown, the curves developed for both types of containers have approximately the same slope.

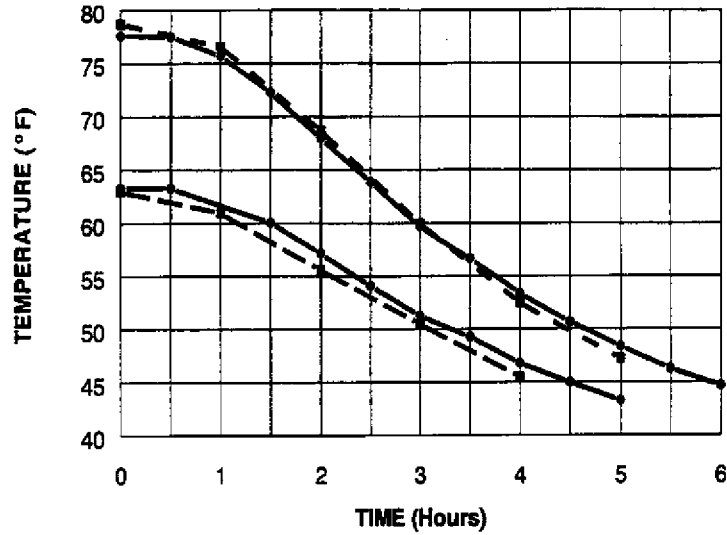


Figure 2. A comparison of the cooling of oyster meats in the geometric center of one-gallon plastic (solid line) and metal (broken line) containers packed in ice. Data for the metal containers (610x708 can) were taken from National shellfish Sanitation Program, Manual of Operations, Part II, Appendix C, page 2.

Cooling curves for solid and wet packs with initial temperatures of approximately 15.6°C (60°F) are compared in Figure 3. The curves are similar which indicates that the addition of up to 25% water to a pack does not change the cooling rate of the oyster meats in the center of container.

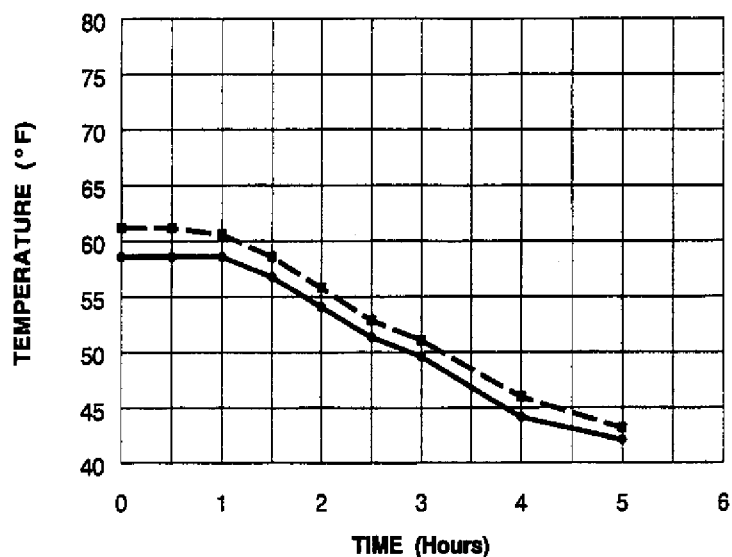


Figure 3. A comparison of the cooling of oyster meats in the geometric center of one-gallon plastic containers with and without added water. Solid pack oysters (solid line) were drained for two minutes before packing. Wet pack oysters (broken line) contained 75% oyster meat and 25% added water by weight.

SUMMARY

The cooling rate of shucked oyster meats in one-gallon plastic containers was approximately the same as that reported for one-gallon metal containers. Temperature at the geometric center of the one-gallon container did not begin to decrease until 0.5 to 1 hour after the container was packed in ice; the rate of decrease thereafter was 2.8°C (5°F) or less per hour. We were unable to achieve an internal temperature of 7.2°C (45°F) at the center of the container within the required two hours with the exception of meats that had been prechilled to less than 10°C (50°F) before packing into gallon containers and icing. Therefore, shellfish processors should consider prechilling shellfish before packing. Strategies for prechilling meats include adding ice to the shucking bucket, using chilled water to wash meats on the skimmer and adding cold water or crushed ice to the tank during blowing.

REFERENCES

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SURVIVAL OF FLORIDA AQUACULTURED CLAMS IN REFRIGERATED STORAGE

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The northern hard clam, Mercenaria mercenaria, is thriving as an aquacultured product of Florida. About 200 shellfish growers currently farm over 700 acres of state-owned submerged lands off two counties on Florida's west coast. Production of hard clams has fast become established in areas where neither aquaculture nor a traditional clam fishery existed. During 1995, sales of clams produced by Florida growers totaled \$5.41 million. This was a 48% increase from 1993. This is equivalent to 43 million live clams sold in 1995. Seeding in 1996 also increased lending to predictions of continued growth for this industry.

The marketability of these clams has been hampered by a perceived short survival in refrigerated temperatures (Menzel, 1972). Commercial hard clams harvested wild in Florida have been both Mercenaria mercenaria and Mercenaria campechiensis and previous work showed that the survival of these clams in common refrigeration decreased significantly as the water temperatures increased throughout the summer months (Otwell et al., 1986; Menzel, 1971).

The objective of this study was to investigate the survival of the Florida aquacultured clam in variable conditions and refrigerated temperatures. Practical methods to increase the survival of the clams and thus improve their marketability were evaluated. These included several tempering techniques which allowed the clams to acclimate to the temperature changes from harvest to storage.

METHODS

This project was initiated by the clam industry in Florida. Farms in four different areas about Florida sent 500 clams from each location (totaling 2000 clams) to the University of Florida, Food Science and Human Nutrition Department the first week of every month for eight months. All the clams were harvested within the same few days and the water temperature, salinity and shipping conditions noted. The target size was 7/8 to 1 inch hinge

width. Immediately post harvest the clams were packaged and sent to Gainesville. Upon arrival at UF, the samples were separated into twelve subsamples. Six of these subsamples were stored in three different temperatures (35, 45 and 55 °F) in two different storage arrangements. Samples were stored in harvest bags to simulate shipment and in trays to simulate retail conditions. Six other subsamples were saved for miscellaneous testing such as freezing, and tempered dry and wet storage with various temperature cooling profiles. In dry storage, refrigerated incubators were programmed to slowly go from harvest water temperatures to storage temperatures. A continuous program (85°F--(20hrs)--45 °F) and a stepwise program (85°F--(6hrs)--65°F--(14hrs)--45 °F) were both investigated. For wet tempering, a recirculating tank was set at 68°F. Clams were placed in the tank immediately post harvest and samples removed after six hours and after 24 hours and placed in the three storage temperatures. All samples were checked daily. The condition of the clams (odor, drip loss and appearance) and the number of dead clams were recorded. If the clams were resting open, they were tapped gently. If they closed they were considered alive. If they remained shut briefly but reopened they were considered commercially dead. If the clams did not shut they were dead. The clams were stored in refrigeration until 50% had died.

RESULTS AND DISCUSSION

The data are presented to show the number of days 80% or more of the clams survived in refrigerated storage from May through October 1996. At the required storage temperature of 45°F the days with 80% clam survival decreased from an average of 17 days in May and June to an average of 8, 7 and 8 days in July, August and September respectively (Figure 1). In October, the average number of days with 80% survival increased up to 16. The survival of the clams from Charlotte Harbor was low in May. That area had been closed for several months due to red tide and this may have weakened and or stressed the clams prior to harvest and shipment. The survival of the clams from Indian River North was low in July following heavy rains which drastically decreased the salinity of the water in that area.

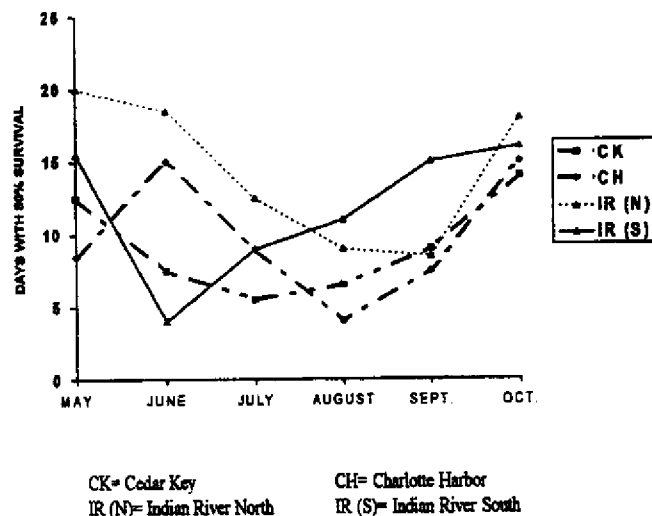


Figure 1. Survival of Florida cultured clams in refrigerated storage (45°F).

The average values for clam survival from all four locations at the three storage temperatures are presented in Figure 2. At 35°F the average days with 80% clam survival was lower than 45°F for all months except May. The greater change from water to storage temperatures stressed the clams further and decreased their survival. As expected the clams stored at 55°F had a longer survival rate than those in the other storage temperatures except in May. Although the clams stored at the higher temperatures lived longer, there were concerns related to the microbiological consequences of storage at the higher temperatures.

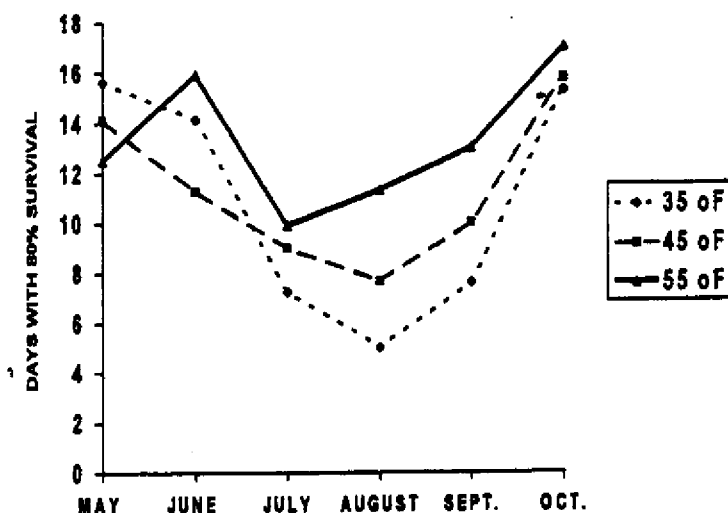


Figure 2. Survival of Florida cultured clams in refrigerated storage.

The water temperatures increased from an average of 76.3 °F in May to an average high of 84.3 in July (Table 1). This approximate 10 degree increase in July and August did have a negative influence on the survival of the clams in all three refrigerated storage temperatures. The salinity of the water at the time of harvest did not appear to influence the survival of the clams but did influence their growth (Table 2). The clams from the areas with the lower salinity were smaller in size throughout the study (Table 3)

Table 1. Water temperatures at harvest for all four locations.

Location	May	June	July	Aug.	Sept.	Oct.
Cedar Key	73	80	86	82	74	69
Charlotte Harbor	80	80	85	85	78	78
Indian River (N)	74	79	84	84	78	72
Indian River (S)	78	80	82	84	80	79

Table 2. Water salinity at harvest for all four locations.

Location	May	June	July	Aug.	Sept.	Oct.
Cedar Key	27	27	27	24	25	22
Charlotte Harbor	32	32	31	32	30	31
Indian River (N)	18	18	13	15	15	15
Indian River (S)	34	32	34	34	32	32

Table 3. Average size (mm) of the clams harvested from all locations.

Location	Cedar Key		Charlotte Harbor		Indian River (N)		Indian River (S)	
	W	L	W	L	W	L	W	L
May	26	47	25	44	20	36	21	39
June	27	48	27	50	20	37	22	40
July	28	49	28	50	22	40	24	41
August	28	48	28	49	24	42	26	42
September	29	50	28	50	22	38	26	44
October	27	48	27	49	19	36	27	45

Tempering the clams from harvest temperatures to storage temperatures in both dry and wet conditions increased the survival significantly. The dry tempering in the refrigerated incubators doubled the number of days with 80% of the clams alive in both July and August, Figure 3. There was no significant difference in clam survival between the continuous and stepwise dry tempers. The number of days with 80% survival of the clams was more than doubled when they were tempered in recirculating seawater, Figure 4. The best tempering results were obtained when the clams were tempered at 68°F for 24 hours post harvest.

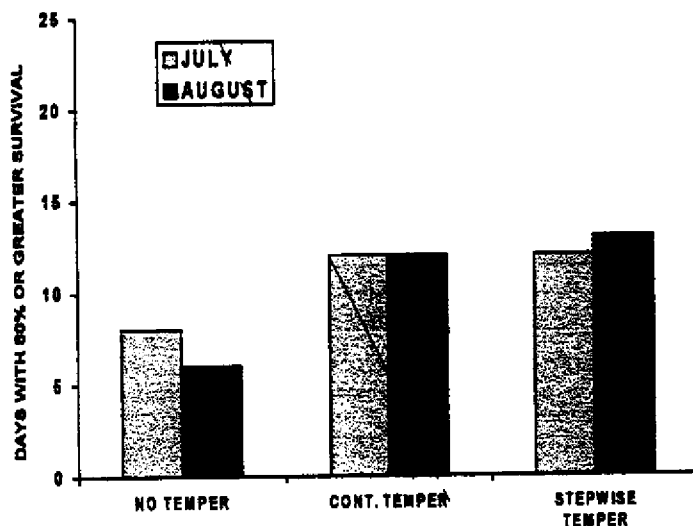


Figure 3. Survival of Florida cultured clams tempered from 85 to 45° F

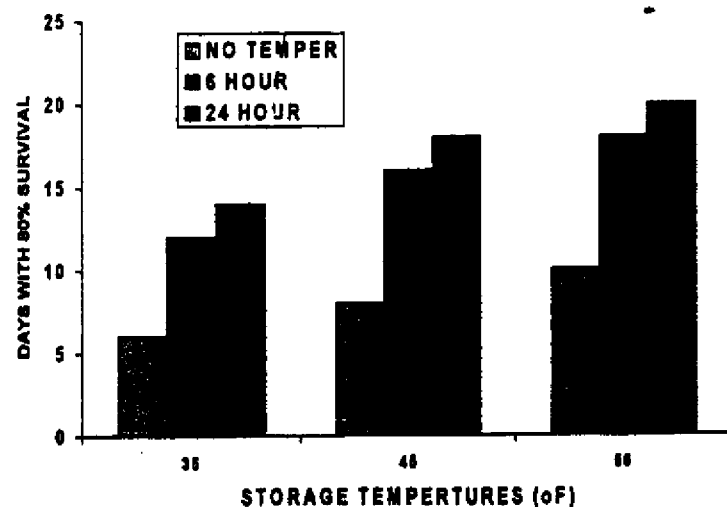


Figure 4. Survival of Florida cultured clams tempered from 85 to 45°F

CONCLUSIONS

The data showed that the primary influences on the survival of clams were the temperatures of the waters at the time of harvest and storage temperatures. When the clams were exposed to rapid and large changes in temperature (>20 degree changes) they became stressed and died. As the water temperatures increased during the summer months the change in temperature between harvest and storage became greater and the clams had shorter survival times. Tempering in both dry storage and recirculating seawater acclimated the clams to the lower temperatures, reduced stress and significantly increased their survival. More research is needed in this area.

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**THE CHARACTERIZATION OF THE PROTEOLYTIC
ENZYME(S) RESPONSIBLE FOR THE POST
MORTEM SOFTENING OF SILVER
HAKE (*Merluccius bilinearis*)**

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INTRODUCTION

Silver hake is a small gadoid fish of great abundance in the North Atlantic. It has not previously been considered a useful species because of its relatively small size (< 30 cm), softening during iced storage and tissue toughening in frozen storage due to the cross linking induced by the enzymatic formation of formaldehyde. Due to the declining traditional fish stocks, the utilization of this species has increased but the problems associated with processing these fish still exist. A potential end-product for silver hake is surimi. To produce a high quality surimi, only the best quality fish muscle can be used. It has been known for some time that when silver hake is stored on ice for any length of time, the tissue begins to soften and is unacceptable for consumption (Hiltz et al., 1976; Leim and Scott, 1966). Therefore the fish must be processed quickly upon arrival at the plant or on board ship, immediately upon landing. As fresh fish must be used to get firm muscle tissue, the fishermen can only remain at sea for a limited amount of time. The fish must be processed quickly in order to ensure quality of the end product.

It is hypothesized that there may be a slow release of membrane-bound proteolytic enzymes causing subsequent softening of the muscle tissue. If this were true, one may be able to correlate the enzyme proteolytic activity in the sarcoplasmic fluid with textural softening. Because there are many proteolytic enzymes associated with fish, only those that are considered active in the physiological pH range, and intimately associated with the myofibrils were examined.

Cathepsin B is a lysosomal cysteine protease that has a major function in intracellular breakdown (Polgar, 1989). An et al. (1994a) showed that one of the most active enzymes in Pacific whiting fish fillets was cathepsin B. Like silver hake, Pacific whiting softens quickly on iced storage. Calpains are neutral thiol proteases, that require calcium as an activator. In comparison to cathepsins, calpains are high in molecular weight (27 KDal for cathepsin, 210

KDal for calpain). Calpain is an intracellular, non-lysosomal cysteine protease (Polgar, 1989). Calpain's main site of proteolysis in postmortem mammalian muscle is the Z-disc and the proteins associated with it, and is thought to be involved with the turnover of contractile proteins (Koochmarai, 1992). However, fish calpains have been noted to also have an effect on myosin and other contractile proteins (Muramoto et al., 1989).

Using phase contrast microscopy and scanning electron microscopy (SEM), the ultrastructure of the myofibrils were examined to help verify the conclusions acquired from the enzyme/texture relationship.

MATERIALS AND METHODS

All reagents were purchased from Sigma Chemical Company (St.Louis MO), unless otherwise specified. Double distilled deionized water was used and all glassware was cleaned in Decon 9, rinsed three times in warm water, and rinsed three times in double distilled, deionized water.

Collection of Samples

Fish were caught, sacrificed and stored on ice in a 30C for not more than three days prior to examination. Fish were randomly picked for each experiment.

Preparation of Crude Sarcoplasm

For all extractions, the sample weights, the volumes of sarcoplasm and the sarcoplasmic protein concentration were measured. Total proteolytic enzymes, calpains and cathepsins were extracted in a one step extraction, described by Nilsson and Ekstrand (1994) with some minor changes. Muscle tissue was excised from the dorsal area of the fish and ultracentrifuged using a Beckman SW-27 rotor, at 100,000 x g for 60 minutes at 3°C. The supernatant was then collected and assayed for protein and enzyme activities.

Enzyme Assays

Optimal pH's and temperatures were determined for all enzyme assays. Temperatures ranging from 0°C - 50°C were tested along with pH levels ranging from 3-8. Calculation of total enzyme present was calculated for each substrate. In order to determine which enzymes were membrane-bound, some samples of muscle tissue were homogenized using a mortar and pestle and centrifuged at 100,000 x g (in order to disrupt lysosomes). The activities were compared to the more gentle treatment of centrifugation which was intended to leave lysosomes intact (Lehninger, Nelson and Cox, 1993). This procedure helped determine which enzymes were present in the sarcoplasm (Nilsson and Ekstrand, 1994). All enzyme extracts were kept on ice until their addition to the reaction mixtures.

Casein Assay

The protocol for apparent calpain activity was described in Wang et al. (1993). Each reaction mixture (in a final volume of 1.0 mL) contained 4 mg of casein, 50 mM imidazole-HCl buffer (pH 7.5) containing 10 mM b-mercaptoethanol, 0.5 mL sodium azide, 0.05 M calcium chloride. To this solution, 1.0 mL of diluted sarcoplasm was added. The samples were incubated for a total of 60 minutes at pH's ranging from 3.5-8 and temperatures ranging from 0°C - 50°C. Once the optimal pH and temperature was established, all other assays were conducted at those values. The reactions were terminated by the addition of 0.5 mL of 10% trichloroacetic acid (TCA). The tubes were then placed in 3-5°C cold room overnight and filtered through Whatman #1 filter paper. Using the method of Lowry et al. (1951), the concentration of TCA-soluble peptides in the filtrate was determined using bovine serum albumin (BSA) as the standard on a Philips PU 8800 UV/VIS spectrophotometer. One unit of calpain activity was defined as the amount of enzyme that caused an increase of one absorbance unit at 280 nm after 60 minutes incubation at 25°C, and corrected by subtracting the activity of a blank that was measured in the presence of EDTA.

Z-Arg-Arg-NMec Assay

The apparent activity of cathepsin B was measured using the procedures described in Yamashita and Konagaya (1990) and Barrett and Kirschke (1981). The muscle was excised as before. The artificial substrate that was used for cathepsin B was Z-Arg-Arg-MCA (benzyloxycarbonyl-Arg-Arg-7-(4methyl) coumarlamide). The samples were assayed at pH's ranging from 3.5-8 and temperatures ranging from 0°C - 50°C. Once the optimal pH and temperature was established, all other assays were conducted at those values. For this assay, 10 mM of Z-Arg-Arg-MCA was used as the substrate. Stock substrate was prepared by dissolving 1mM Z-Arg-Arg-MCA in sulphonate, and kept at 3°C until needed and made 20 mM with 1% Brij solution daily.

The activities were measured by mixing 1 ml of the appropriately diluted amount of muscle extract with 1% Brij solution, 1 mL of 0.2 M citric acid-phosphate buffer (0.1% b-mercaptoethanol, prepared daily), waiting one minute for activation and then beginning the reaction with the addition of 1 mL of 2 mM substrate (Z-Arg-Arg-MCA). Samples were incubated at the appropriate temperature and pH, using a temperature controlled cell in a luminescence spectrophotometer (Perkin Elmer C550 Luminescence Spectrophotometer), set at excitation 370 nm, and emission 460 nm. A standard of 7-(4-methyl) coumarlamide was made to quantitatively measure the amount of product being liberated. This reaction was performed in the dark because 7-(4-methyl) coumarlamide is photo-reactive. One unit of activity was expressed as that releasing 1 mmol of aminomethylcoumarin/min at 25°C.

Inhibition/Activation Assays

Inhibitory assays were performed to confirm the identity of the enzyme(s) suspected to be the cause of the softening. In each case, the same enzymatic assay was performed both

with and without the inhibitor or activator. Refer to Table 1 for the effect of inhibitors and activators used.

Table 1. Common protease inhibitors/activators and their effects

Inhibitor	Effect
E-64*	Cysteine protease inhibitor, blocking thiol groups.
CaCl ₂	Essential for the activity of calcium activated proteases.
Trypsin Inhibitor	Serine protease inhibitor.
EDTA	Chelates divalent cations such as Ca ²⁺ .
Leupeptin	Cysteine protease inhibitor, blocking thiol groups.
Antipain	Cysteine protease inhibitor.
Pepstatin	Carboxyl protease inhibitor.
Iodoacetic acid	Cysteine protease inhibitor

* E-64 (L-trans-epoxysuccinylleucylamido(4-guanidino)butane.

Inhibitor and activator concentrations were described in Yamashita and Konagaya (1992) and Kominami et al. (1984). The concentrations of each inhibitor used in each assay were 2 mM E-64, 50mM CaCl₂, 0.05% soy trypsin inhibitor, 2mM EDTA, 0.1 M leupeptin, 10 mg/mL antipain, 0.1 M pepstatin and 2 mM iodoacetic acid. Calcium chloride is an activator for calpain like enzymes, and was omitted from casein assay to show its affect.

Texture Analysis

Texture analysis was performed using an Instron Model 4502 (Instron, Canton, MA) equipped with a Kramer shear-compression cell (Gill et al., 1979). Fresh uncooked fish were used for texture measurements. The Instron Series IX Automated Materials Testing System (Version 5.02, Instron Corporation) was used to calculate the shear force and peak force at various user defined points on the texture profile (Figure 1).

Phase Contract Microscopy

Aseptically dissected muscle tissue (10g) and 90g of distilled water was homogenized in a stomacher (Lab Blender 400) for 30 seconds in order to prepare a fine suspension of individual myofibrils. A drop of this homogenate was placed on a clean microscope slide with a cover slip and viewed at 400x under oil immersion on a phase contrast microscope (Nikon Optiphot) equipped with a Nikon (FX-35A) camera.

Electron Microscopy

Samples of fish fillets (three) were wrapped in plastic, iced and transferred immediately to the electron microscopy laboratory of the "Institute for Marine Biosciences" (National Research Council of Canada, Halifax, NS). The fillets were further dissected into 1 mm

samples, ensuring that for each sample, there was a longitudinal section, an oblique section, and a transverse section. These samples were plunged into liquid propane, cooled near liquid nitrogen temperatures. These samples were fractured by striking with a sharp blade, freeze dried in a Meitech Model 750 freeze drier and gold-coated in an Edwards 306A coater. The samples were examined using a scanning electron microscope JEOL model JXA35 at magnifications of 2000X and 4000X.

Statistical Analysis

Systat version 5.05 for Windows was used to analyze data using ANOVA stepwise regression, calculating the t-value and F-value for a 5% level of significance. Error bars on figures represent 2 standard deviations.

RESULTS AND DISCUSSION

Texture Analysis

Ando et al. (1991) used a puncture test to show that the firmness of muscle tissue during iced storage had a good correlation with sensory results. Although this study was used to show a relationship between the softness of sashimi (a raw fish meat dish) and the perceived mouth feel, the study also showed that the break strength of muscle tissue correlated with tissue softening.

After several trials, it became qualitatively apparent that the modified Kramer cell force deformation curves gave reasonable measurements of softening of silver hake muscle tissue. Several portions of the force deformation curves were examined. Of the 5 points measured, the maximum load and firmness (Figure 1) best measured the softening of the silver hake. Because of the size of the fish, a pooled sample was used for analysis. Therefore any variation within each test period was test variation, not sample variation. Figure 2 shows that during iced storage, the muscle tissue became softer and after 6 days of iced storage, developed a "pudding-like" texture.

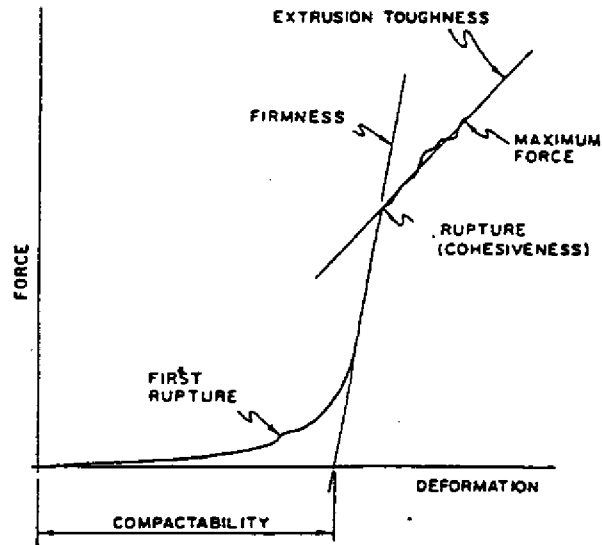


Figure 1. Typical force deformation curve, illustrating possible points of significance (Voisey and Larmond, 1977).

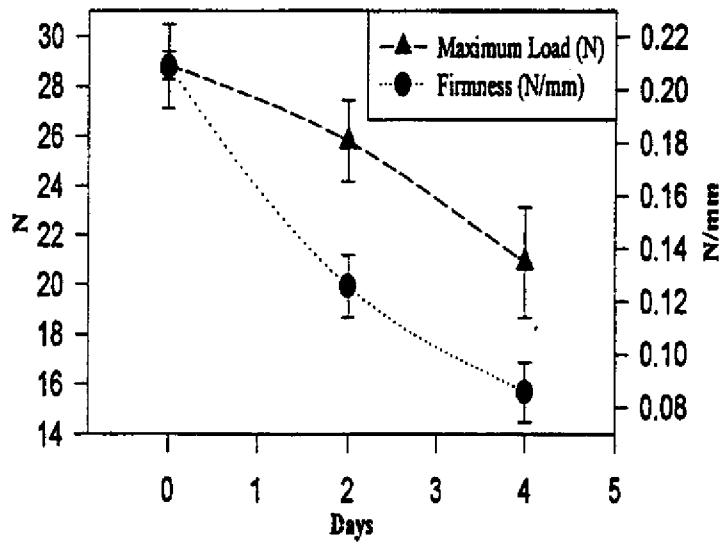


Figure 2. Texture profile analysis of silver hake stored on ice for 4 days ($n=4$, Max. Load, $T=7.084$, $F=50.84$, $p=0.000$; Firmness, $T=10.388$, $F=1-7.906$, $p=0.000$). Refer to Figure 1 for definition of points.

Casein Assay

Apparent millimolar calpain activity was monitored using the method of Wang et al. (1993) using casein as a substrate. Casein hydrolyzing activity was inhibited by E-64 (99.8%), EDTA (85%), leupeptin (91%), antipain (92%) and IAA (88%). Caseinolytic activity was enhanced by the addition of Ca^{+2} . The optimum pH was higher than the pH of the fish muscle extract, but there was activity between pH 6 and 6.5. The optimal pH was measured to be 7.1, with an optimal temperature of 30°C. There was an increase in the specific calpain-like activity in the sarcoplasm over time of iced storage (refer to Figure 3). It is speculated that mechanism for the softening of the muscle tissue may not be one agent but rather a combination of several enzymes. It would appear that calpain was one of the enzymes that had an affect on the overall texture of silver hake muscle tissue.

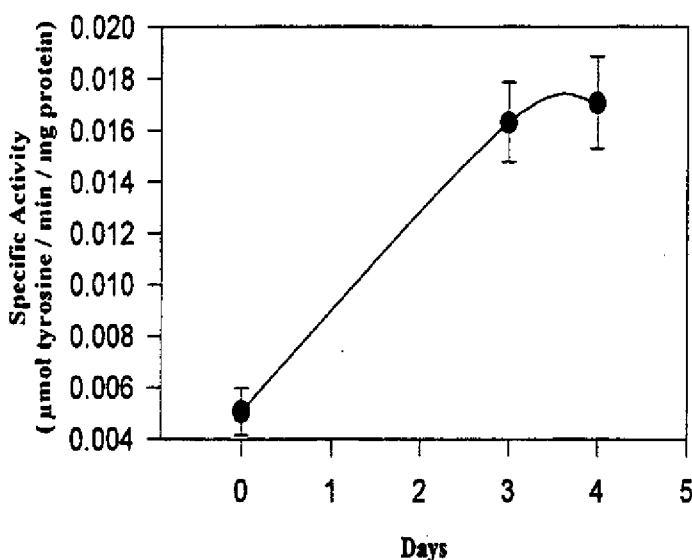


Figure 3. Casein-hydrolysing activity from silver hake tissue extract, pH 7.1, Temp 30°C (n=6, T=11.512, F=132.532, p=0.000).

Z-Arg-Arg-NMec Assay

It was expected that the cathepsins may have had a greater influence over the softening of the muscle tissue than the calpains although the latter are very powerful proteolytic enzymes and thought to be the causative agent in the softening of mammalian muscle tissue (Koomaraie, 1992). The fluorescence of aminomethylcoumarin liberated from the substrate was measured by excitation at 370 nm and emission at 440 nm with a fluorescence spectrophotometer. The cathepsin B specific activity increased nearly 5-fold over time of iced storage (Figure 4). The total cathepsin B (bound plus unbound activity) was also calculated and showed that 96% of the cathepsin B was solubilized during iced storage. All assays were completed at the optimal pH determined to be 6.5, with an optimal temperature of 27°C. Cathepsin B has been regarded as the most active cysteine protease in Pacific whiting fish fillets (An et al., 1994b). Because silver hake is a close relative of Pacific

whiting, it would be reasonable to assume that cathepsin B would be a very important factor in the proteolysis of silver hake muscle tissue.

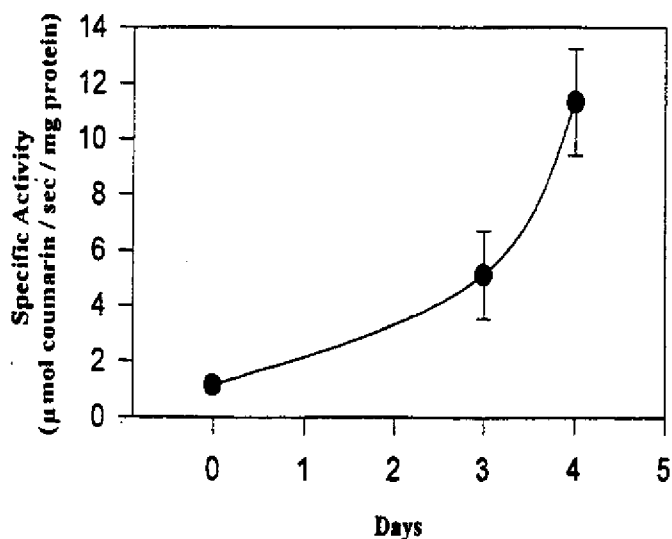


Figure 4. Z-Arg-Arg-NMec hydrolysing activity from silver hake tissue extract, pH 6.5, temperature 25°C (n=6, T=6.744, F=45.481, p=0.000).

Inhibitors and Activators

Inhibition tests were conducted to help with the identification of each enzyme and to eliminate the possibility of others (Table 2). The inhibitors used were E-64, pepstatin, EDTA, trypsin inhibitor, leupeptin, antipain, calcium chloride and iodoacetic acid. The caseinolytic activity results suggest the enzyme was a calcium-activated cysteine protease, as illustrated by its decrease in activity with E-64, leupeptin, antipain and iodoacetic acid EDTA. CaCl₂ was obviously an activator for the caseinolytic activity. The minimum decrease in activity with trypsin inhibitor, and pepstatin would also help confirm that most of proteolytic activity was not a trypsin like enzyme or a carboxyl protease. Cathepsin B which was assayed in the presence of a rather specific artificial substrate (Z-Arg-Arg-NMec) was not affected by the presence of Ca²⁺, pepstatin or EDTA, but was inhibited in the presence of E-64, leupeptin, antipain and iodoacetic acid.

Table 2. Results of the effects of different inhibitors and activators on the activity of silver hake sarcoplasm (percentage of activity with inhibitors present in assay)

Inhibitor	Cathepsin B	Calpain
E-64	0.6	0.2
Pepstatin	93	86
EDTA	96	15
STI	87	89
Leupeptin	12	9
Antipain	17	8
CaCl ₂	96	130
IAA	6	12

Texture and Enzyme Activity

For each of the enzymes assayed, the muscle texture was analyzed along with the specific activities of apparent calpain and cathepsin B. It is difficult to establish a "cause and effect" relationship for such a limited set of data. However, Figure 5 shows the results for casein hydrolyzing activity versus the texture (firmness) and it can be seen that there was negative relationship. That is, softening generally corresponded to higher specific activities. This would indicate that as the casein hydrolyzing activity increased, the texture of the muscle tissue became less firm. The same can be seen in Figure 6 with regard to the Z-Arg-Arg-NMec hydrolyzing activity. It should be noted that no error bars are present and ANOVA was not performed because two dependent variables are being compared.

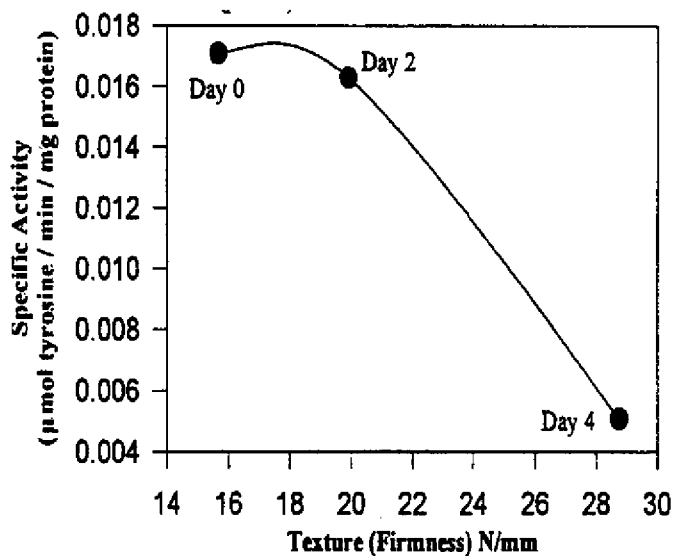


Figure 5. Relationship between texture (firmness) of silver hake muscle tissue and casein hydrolyzing activity of sarcoplasmic fluid (pH 6.3).

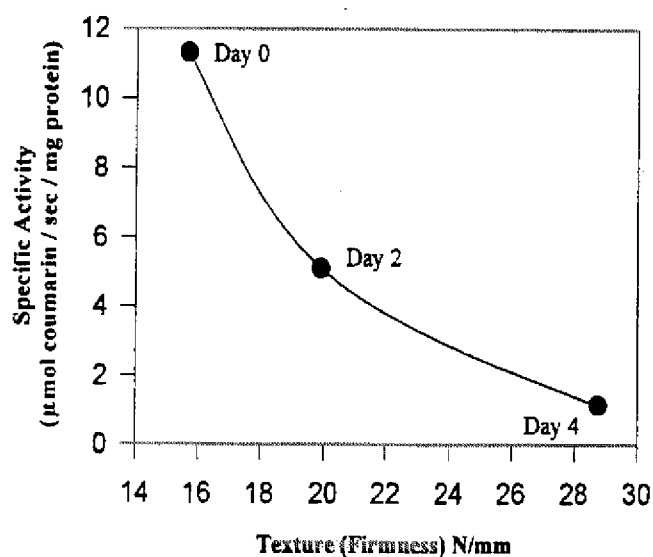


Figure 6. Relationship between texture (firmness) of silver hake muscle tissue and Z-Arg-Arg-NMec hydrolysing activity of sarcoplasmic fluid (pH 6.3)

Phase Contract Microscopy

There was a reduction in the length of myofibrils over time on iced storage. Similar observations were made by Tokiwa and Matsumiya (1969) who were studying cod, pollack and carp at the time. The fresh silver hake myofibril strands were long and thread-like. As time on iced storage increased, there was an increase in the fragmentation and a decrease in length of the myofibrils (Figures 7 and 8). This agrees with observations seen on the myofibrillar fragmentation of stored cod muscle tissue (Tokiwa and Matsumiya, 1969).

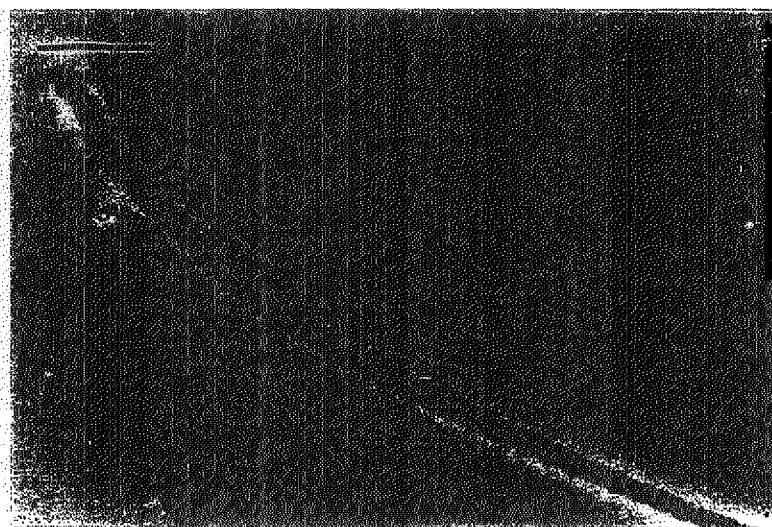


Figure 7. Fresh silver hake myofibrils viewed using a phase contrast microscope under oil immersion (bar = 10μm)

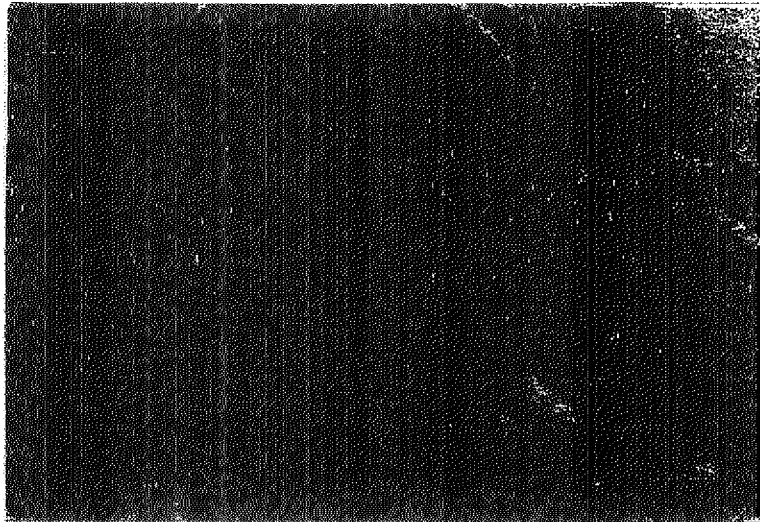


Figure 8. Silver hake myofibrils after 6 days of iced storage viewed using a phase contrast microscope under oil immersion (bar = 10 μ m).

The typical striated appearance of the myofibrils changed with iced storage time. The structural definition of the Z-line decreased with time on ice, suggesting degradation of the contractile proteins. Tokiwa and Matsumiya (1969) also observed that the Z-line deteriorates during iced storage, which would make the muscle tissue more susceptible to mechanical breakage. They speculated that because of the optimal pH of the protease in question and of the hydrolyzed proteins, that the enzymes most likely responsible for this fragmentation were cathepsins. The activities of both cathepsin B and calpain had the Z-line as a potential substrate.

Scanning Electron Microscopy

Scanning electron microscopy was used to see the ultrastructural changes. Figure 9a shows a cross section of a myofibril. The myofibrillar protein network that was broken down in the iced silver hake (Figure 9b). Figure 10a shows a longitudinal view of a myofibril. Again the inter-connection present in fresh muscle tissue deteriorates during iced storage (Figure 10b). Not only was there a reduction in the proteinaceous network seen in myofibrils but also in the collagen sheath surrounding the fibre. Bremner and Hallet (1985) also reported the degradation of collagen in fish muscle during storage. The reduction in the collagen sheath surrounding the fibre could explain the mechanical breakage occurring during fragmentation of tissue samples.

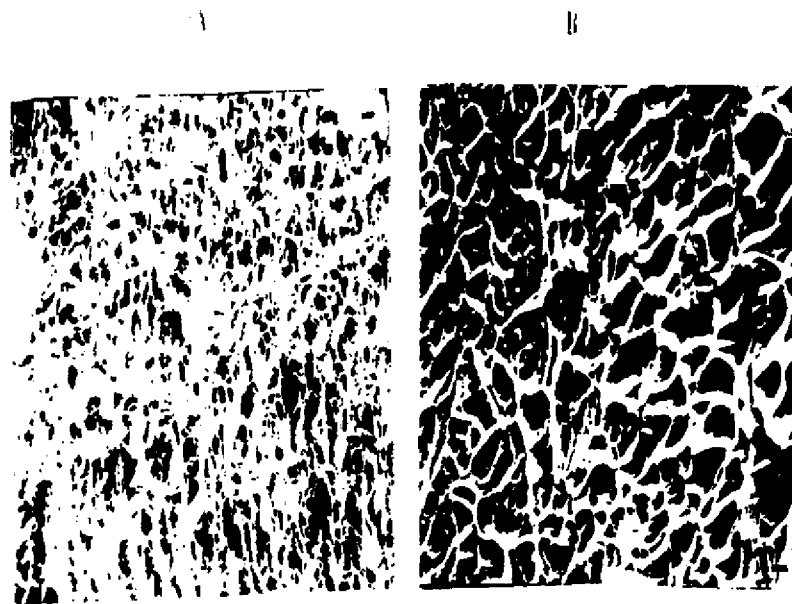


Figure 9. Silver hake myofibrils (transverse section) viewed using a scanning electron microscope a) Fresh myofibrils b) Myofibrils stored on ice for 6 days (Bar = 10 μ m).



Figure 10. Silver hake myofibrils (longitudinal section) viewed using a scanning electron microscope a) Fresh myofibrils b) Myofibrils stored on ice for 6 days (Bar = 10 μ m).

CONCLUSIONS

Both cathepsin B and calpain are capable of hydrolysis under physiological conditions. Both of the enzymes tentatively identified in silver hake had specific activities which increased as time on iced storage increased. The inhibition studies showed that one enzyme was most likely a cysteine protease, and not a trypsin-like protease or a carboxyl protease. The second enzyme was more typically like cathepsin B, cleaving the artificial substrate, Z-Arg-Arg-NMec and inhibited by E64, leupeptin, antipain and iodoacetic acid. The texture of the muscle tissue became very soft after only 4 days of iced storage. The microstructure and ultrastructure of the muscle tissue showed degradation of the contractile proteins of the myofibril. The relationship between the texture softening and the activities of the two substrates indicate that the two most likely enzymes causing the softening of the silver hake muscle tissue were calpain and cathepsin B.

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THE VOLUNTARY NUTRITION LABELING OF RAW PRODUCE AND FISH: FDA'S POLICY ON DATA BASE REVIEW

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On August 16, 1996, the Food and Drug Administration (FDA) published a final rule that revised the guidelines and the nutrition labeling values for the voluntary nutrition labeling of raw fruits, vegetables, and fish. The action, in response to the requirements of the Nutrition Labeling and Education Act of 1990 (NLEA), makes the voluntary nutrition labeling program more consistent with mandatory nutrition labeling of other foods regulated by FDA. Within the same document, the agency also explained its policy on its review of databases in both the voluntary and mandatory nutrition labeling programs.

In November, 1991, FDA published a final regulation in the Federal Register (corrected March, 1992) that:

- 1) identified the 20 most frequently consumed raw fruits, vegetables, and fish in the United States;
- 2) established guidelines for their voluntary nutrition labeling; and
- 3) set criteria for substantial compliance with the guidelines by food retailers.

The agency also stated that at least every two years it would publish updates on the values and provide an opportunity for comment. Otherwise, FDA would publish a notice that nutrition labeling values had not changed. [The 1996 regulation increased that time interval to every four years.] FDA also advised that once final regulations governing nutrition labeling of FDA-regulated processed, packaged foods were finalized (in 1993), it would revise the guidelines for the voluntary nutrition labeling program to make them as consistent as possible with those final rules. Consistency is defined in terms of what information is required (content), and how that information is to be presented (format).

FDA published a proposal in the Federal Register in July, 1994, to update the nutrition labeling values for the 20 most frequently consumed raw fruits, vegetables, and fish and to revise the guidelines for the voluntary nutrition labeling of these foods to reflect the final rules for mandatory nutrition labeling. The agency considered all comments to the proposal in finalizing the regulation.

Definition of "Raw Fish"

FDA considers "raw fish" to be freshwater or marine finfish, crustaceans, and mollusks in the natural state that have received minimal or no processing. Raw fish may include whole or filleted fish that are fresh or fresh frozen (unpacked or packaged by the retailer); alive in the retail store; shrimp that have been shelled or deveined; and lobster, crab, and shrimp that have been thermally processed or shelled, but not processed or prepared in any other way. Nutrition labeling is mandatory for fish that are canned or smoked; have undergone processing such as breading, flaking, or pressing; or were packaged before reaching the retail level. FDA doesn't recommend that consumers eat raw fish, however, so the agency reports the nutrient values on a 3 oz. skinless, cooked portion, without added, salt or sauces.

The 20 Most Frequently Consumed Raw Fish

Blue Crab	Orange Roughy	Swordfish
Catfish	Oysters	Whiting
Clams	Pollock	
Cod	Rainbow Trout	
Flounder/Sole	Rockfish	
Haddock	Salmon (Atlantic/Coho)	
Halibut	Salmon (Chum/Pink)	
Lobster	Salmon (Sockeye)	
Mackerel	Scallops	
Ocean Perch	Shrimp	

The Guidelines

The final regulations for the voluntary nutrition labeling program continue to grant retailers flexibility in disseminating the nutrition labeling information to consumers through various means and materials, such as shelf labels, signs, posters, brochures, notebooks, or leaflets. Much of the regulation does relate to retailers, but it also relates to members of the industry who wish to label individual products and to those who want accurate nutrient levels and label values for fish.

Retailers may use a chart format or an individual label format, as long as the labeling materials are viewable from within the fish department. In addition, if raw fish are frozen within the retail establishment and presented in a different section of the store, that section should also provide the labeling information. Many of the trade associations are continuing to use a chart format in their marketing materials, but FDA encourages industry to consider using an individual label format, the Nutrition Facts panel that you see on processed, packaged foods. A poster or brochure for fish could easily contain 20 individual nutrition

labels and may be easier to read than a chart format with 20 lines and columns that contain the nutrition information.

The new regulation requires that retailers, as well as members of the fish industry who wish to provide individual nutrition labels on packaging materials for fish included in the voluntary program, should use the nutrition labeling values provided by FDA for the most frequently consumed raw fruits, vegetables, and fish. After August 16, 1997, the effective date of the regulation, retailers must use the data provided by FDA in order to be in compliance with the guidelines for the voluntary nutrition labeling program. That date is also the effective date for members of the fish industry who label individual packages of fish.

FDA recommends that labeling values be used as soon as possible, however, especially at the retail level. Because there is a relatively short amount of time before the 1996 FDA compliance survey, FDA will consider either the old (1991) or new (1996) labeling values for retail stores to be in compliance.

Individually labeled fish products will not be assessed for compliance as a part of the 1996 FDA compliance survey for the voluntary nutrition labeling program, but they will need to be in compliance with the regulations for mandatory labeling in § 101.9. Those regulations explain the requirements for the Nutrition Facts label in regard to format, type size, bold print, etc. FDA does plan to assess the prevalence of individually packaged raw foods that bear nutrient content claims, such as "low fat" and whether or not they have the nutrition facts panel.

Nutrient Levels

FDA calculated the labeling values for raw fish based upon raw data obtained from the United States Department of Agriculture (USDA). The agency based values for catfish on data in the *Journal of Food Science* in a 1990 article by Joyce Nettleton, William Allen, and several other authors. Those data were provided to FDA in a comment to the proposed rule.

Again, FDA analyzes raw nutrient data. FDA believes that mean values from software data bases or the scientific literature may be inappropriate for nutrition labeling and does not recommend their use except for calculating nutrients for restaurant menus. The agency derives labeling values by completing compliance calculations with the data, using 95% prediction intervals. The final labeling values will be adjusted from the mean that would be directly calculated from the data. Sometimes the levels of "bad" nutrients will be inflated; other times the levels of "good" nutrients will be deflated. Rather than using a mean value alone, FDA looks at all data and considers the variability among the data points in calculating a label value for a nutrient.

FDA strongly encourages the fish industry, trade associations, and academia to continue to test fish to determine nutrient levels and to provide those data (especially raw data) to the agency for consideration in the next update of labeling values.

The regulation states that in four years, after reevaluating the most frequently consumed fish in the United States, the agency will provide an update to the regulation. For example, there is already a question whether mackerel should be included on the list. The agency plans to continue to work with the National Fisheries Institute and other interested parties to continue to refine the list of fish. FDA strongly encourages you to send not only data for nutrient values for fish but also estimates to determine current consumption of various types of fish. Please remember that consumption is not the same as sales or catch.

FDA requires certain nutrients for food labeling (i.e., calories, calories from fat, total fat, saturated fat, cholesterol, sodium, total carbohydrate, dietary fiber, sugars, protein, vitamin A, vitamin C, calcium and iron) but also allows for optional nutrients for foods in the voluntary program. Even though potassium is optional, FDA notes that the potassium provided by fish is important--12 out of the 20 types of fish are considered a "source" of potassium--so the agency included potassium in its data. The individual label format provides an easier vehicle to list optional nutrients.

The regulations state that on charts the columns for sugars and fiber may be omitted for fish. Instead, a footnote may be included, stating "Fish provide negligible amounts of dietary fiber and sugars." With fewer columns, the charts would be more readable by consumers, but the amount of information provided would not be reduced.

Nutrient Content Claims

Nutrient content claims are label statements such as "low fat", "good source of potassium". FDA addressed the question of the need for nutrition labeling for packaged raw fruits and vegetables that bear a claim in a booklet on frequently asked questions that it issued in August of 1993. At that time, it stated: "Claims subject the food to nutrition labeling in accordance with § 101.45, which means that nutrition information will have to be available at point of purchase although not necessarily on the package." The agency is now reevaluating that policy for not only raw produce, but for raw fish.

FDA encourages members of the fish industry who put nutrient content or health claims on their packaging to also include nutrition label information because it is not possible to predict whether the products will be sold in stores where retailers make the nutrition information available to consumers. Even if it is possible to control the flow of the products into specific retail stores, it is not possible to have control over retailers' decisions to display (or to continue to display) nutrition labeling information for these products. Depending upon retailers to provide nutrition labeling values to justify nutrient content or health claims would be a gamble for those who assume liability for their products with claims. For raw fish that are not among the 20 most frequently consumed, it is even less likely that nutrition information will be available in retail stores. Therefore, FDA recommends that nutrition information be included on those products bearing claims.

Submitting Data to FDA

The nutrition labeling values provided by FDA are for generic commodities. Several groups have called to ask if a commodity group could use nutrition labeling values developed for a specific type of fish. FDA does encourage the use of names and label values for specific products, as long as the commodity group has the data to support the label values used for the product. If a commodity group wishes to amend the nutrient values for a generic item, FDA encourages the group to submit the values to the agency for consideration in the agency's next revision of the voluntary nutrition labeling program, which would be in 2000. Of course, you'd want to submit the data prior to that time. In order to log a request into the system, please send all submissions to:

Food and Drug Administration
Center for Food Safety and Applied Nutrition
Office of Food Labeling, (HFS-165)
200 C St., SW
Washington, D.C. 20204

If you have questions, please call Mary Bender 202-205-5592; send a fax to 202-205-5532; or best yet, send an E-mail to m0b@fdacf.ssw.dhhs.gov. [That's m-zero-b.]

If FDA decides to use the labeling values you submit for the generic item, those values will be made available for public comment. Again, any nutrition labeling value for a generic item that the agency decides to incorporate will have to be used by retailers for them to be in compliance.

Issues on Raw Fish

Catfish. There were three comments to the proposed rule that dealt with the fat content of farmed catfish. I understand that the fat content is affected by species, size of fish, diet of fish, season of year, stocking rate of pond, pond size, and gender of fish. The comments also provided data from Nettleton et al. on farmed catfish composition and requested that FDA consider this information in developing revised labeling values.

FDA concluded that the data from Nettleton et al. provided more accurate label values for catfish. So the agency derived values and adopted them. The values for total fat are higher than the industry wants, however, and the industry is concerned. Again, we encourage all to continue to analyze data for raw fish and to submit those data to FDA.

Orange Roughy. FDA is still concerned about the fat value for orange roughy, because the current value for orange roughy doesn't include wax esters and is not consistent with the definition of total fat in the regulations (i.e., the amount of total lipid fatty acids present expressed as triglycerides).

A comment to the proposed rule did question why FDA wanted a value for total fat in orange roughy that includes the presence of wax esters because wax esters are not a

metabolizable source of energy in humans and have no dietary significance. It also stated that consumers need information with which to make dietary choices, and that it is misleading to add nonmetabolizable fat to the value for fat in orange roughy. The elevated levels of fat that would result from the addition of wax esters would falsely suggest to consumers that orange roughy was contributing a substantial amount of metabolizable fat to daily intake.

FDA continues to request information that would provide a basis for revising the declaration of total fat to reflect the presence of wax esters in orange roughy but that would not be misleading to the consumer. The agency will also address the issue of declaration of available fat in a separate rulemaking.

Atlantic/Pacific Mackerel, Ocean Perch, and Lobster. In response to comments to the proposed rule, FDA reviewed the data provided to the agency for mackerel and ocean perch and discovered that there were errors in the data file. FDA obtained the correct information and then derived the nutrition labeling values correctly for the final rule. Thanks to the commenters, FDA was able to trace the data errors to their source and make the corrections. In addition, the agency did make an error and correct it in the final rule in regard to calories from fat for lobster.

FDA Review of Submitted Data Bases

FDA needed to create a more efficient, flexible and responsive data base review system that wouldn't overwhelm the resources that the agency has available, and yet provide industry with the assurance that it seeks through data base review and approval. FDA solicited comments regarding the agency's approach to data bases in the proposal on the voluntary labeling of raw produce and fish. Based on its review of the comments, FDA decided to modify its approach to data bases that are submitted to the agency for review. The new policy relates to products falling under the mandatory label program, but also includes foods in the voluntary program, such as raw fish.

All data in the form of nutrition label values that are submitted to the agency should be accompanied by raw data. If there are data that the submitter has determined as unsuitable, they should also be provided with explanation.

FDA will continue to evaluate data submitted for the 20 most frequently consumed raw fish. In addition, FDA will evaluate data for fish NOT included in the top 20 that are submitted for review if those data are accompanied by a plan to collect additional data for the purpose of updating label values. [In other words, if you want to send data for the top 20, please do. If you want to send data for other types of fish, please do, as long as you also submit a proposal to collect additional data.]

In order to facilitate the use of the developing nutrient data base for fish and to limit the uncertainty that could result from a delay in agency review of the data base, upon submission firms may begin use of the nutrient label values and to initiate the planned studies to collect

data to update the values. During this interim period, FDA won't take action against a product bearing label values included in a data base submitted to the agency for review. If any product is identified through FDA compliance activities as including label values that are out of compliance, contingent on the company's willingness to come into compliance, the agency will work with both the manufacturer and the data base developer to understand and correct the problematic label values.

When FDA receives the interim data and planned studies, it will first evaluate the label values relative to the raw data. FDA will recalculate label values based solely on the raw data that have been submitted. As explained earlier, the agency will derive label values using compliance calculations based upon 95 percent prediction intervals and, when appropriate, will use weighting procedures, as recommended in the FDA Nutrition Labeling Manual.

FDA will evaluate the data for completeness and reasonableness, e.g., it will consider whether or not there are enough samples, and whether all nutrients are included. FDA requests that supporting documentation, such as analytical methodology and a sampling plan, accompany interim data. The agency acknowledges, however, that a large amount of the interim data available from manufacturers and trade associations are based upon historical data, where the analytical methodology and sampling plan are not available. Therefore, FDA will accept data even if it is not accompanied by comprehensive documentation, as long as the reason that the documentation is not provided is fully explained and is acceptable to the agency.

After FDA reviews the data, it will consider using those values in updating the top 20. For fish not included in the top 20, FDA will review the planned studies to collect additional data. The agency will concentrate on analytical methodology and on the reasonableness of the factors that could account for nutrient variability (e.g., region), rather than on the rigor of sampling design or statistical treatment of the data. FDA cautions, however, that data base submitters should follow the FDA recommendations regarding sampling strategies, weighting procedures, and statistical treatment of data that are described in the nutrition labeling manual.

FDA will respond in writing after review of the data and the planned studies. The agency will address the nutrient label values that were submitted and will indicate whether it has any objection to continuing the planned studies or to continued use of the label values for two years from the date of the agency response. After those two years, manufacturers will be expected to provide the agency with a summary update that reassesses the interim label values based upon completion of the planned laboratory analyses. The agency will evaluate how the study findings bear on the interim label values and will consider whether it would have any objection to continued use of the updated interim values for up to an additional five years. At the same time, however, the agency may suggest modifications to the ongoing plan of study. If after review of data and planned studies, FDA determines that the label values or studies are not appropriate, as indicated above, the agency will notify the manufacturer of that decision.

Please note that an initial primary focus of FDA's compliance review of product labels is on nutrient content claims (e.g., "high protein", "low fat") that are used. FDA will continue to closely monitor products making such claims and expects that the manufacturer, packer, or distributor will have sufficient data to support the validity of such claims.

Again, FDA strongly encourages industry to analyze data for raw fish and to submit those data to the agency for consideration for the next revision of nutrition labeling information for raw commodities. If an updated rule is to publish in 2000, we'll need data in the next two years in order to consider it in a proposal.

**EXTRACTABILITY OF MYOFIBRILLAR PROTEIN FROM COD
(*Gadus morhua*) FROZEN STORED AT DIFFERENT
TEMPERATURES AND MUSCLE INTEGRITY¹**

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Formation of intermolecular bonds among myofibrillar proteins is the main cause of loss of functionality and texture change during frozen storage of many fish species of high commercial value. When this occurs, it spells the end of practical storage life. In gadoid species like cod (*Gadus morhua*), this aggregation is thought to be caused largely by formaldehyde (FA) forming in the muscle through demethylation of trimethylamine oxide (TMAO) (Matsumoto, 1979; Shenouda, 1980), although the means by which FA causes protein aggregation is not yet clear and the results in the literature are contradictory (Connell, 1965, 1975; Dingle *et al*, 1977; Owusu-Ansah and Hultin, 1987; Ragnarsson and Regenstein, 1989, Hultin, 1992; Del Mazo *et al*, 1994; Tejada *et al*, 1995).

The changes occurring in muscle proteins differ according to frozen storage time and temperature, degree of intactness of the muscle and fish species (Careche *et al*, 1995; Careche *et al*, 1996; Tejada *et al* (in press).

In view of the financial losses consequent upon altered texture and functionality in gadoid fish, many studies have been conducted on proteins in frozen stored muscle in order to ascertain the cause of such aggregation. However, not much work has been done on intact or minced frozen muscle stored at different temperatures under standardised conditions in order to study changes in extractability and solubility of natural actomyosin (NAM) in solutions capable of selectively breaking different protein bonds, and to examine the types of insoluble aggregate generated under different solubilisation conditions.

1. This work is part of a project financed by EC (Project FAR-UP3 647) and Spanish CICYT (ALI 92-1354-CE). Part of the results have been presented in previous meetings.

OBJECT

The aim of this study was to identify the changes occurring in natural actomyosin (NAM) isolated from frozen fillets and minces obtained from cod, with a view to ascertaining the bonds responsible for the formation of protein aggregates during frozen storage at different temperatures or states of muscle intactness.

MATERIALS AND METHODS

Fish source

Frozen cod (*Gadus morhua*) fillets and minces were supplied by CSL Food Science Laboratory, Torry, Aberdeen, UK, air-freighted with solid CO₂. They were then vacuum-packed and stored at -20° C [minces and fillets and at -30° C [fillets]] for up to 62 weeks. The mince was made from fillets taken from 20 individuals, using an Omega TE22 mincer with 5-mm hole size.

Natural actomyosin extraction

Natural actomyosin (NAM) was extracted with 0.6M NaCl from 100 grams of thawed mince or fillets by the method of Kawashima *et al.* (1973). The centrifuging force applied was 5000g (Sorvall RT6000B, DuPont Co., Wilmington, DE). This was fraction S1. Protein concentration in the supernatants was determined by the Lowry method (Lowry *et al.*, 1951; Peterson, 1979) and Kjeldahl (AOAC, 1984).

Extractability of aggregates

The insoluble materials from the 0.6M NaCl (P1) extractions were treated with 4 volumes of 2.5% sodium dodecyl sulphate (SDS) (Merck, Darmstadt, Germany) stirred with a magnetic stirrer for 10 min at room temperature. After centrifugation (Sorvall RT6000B, DuPont) for 15 min at 5000g, it was washed again with 1 volume of 2% SDS and re-centrifuged. This was fraction S2. Any remaining aggregate (P2) was treated with 2% SDS plus 5% β-mercaptoethanol (ME) (Merck, Darmstadt, Germany) as before, to obtain fraction S3 and in some cases an insoluble precipitate (P3). The purpose of these two extractions was to break down non-covalent bonds and the disulfide bond respectively. The amount of soluble protein in fractions S2, S3, and P3 was determined by Kjeldahl (AOAC, 1984).

Polyacrylamide gel electrophoresis

All extracted fractions were analyzed by SDS-PAGE in a Phast-system horizontal apparatus (Pharmacia LKB Biotechnology, Uppsala, Sweden), using 12.5% polyacrylamide gels. Samples were treated according to Hames (1985) (2% SDS, 5% ME, and 0.002% bromophenol blue (Merck) and then heated for 5 min in a boiling water bath. Samples were

then centrifuged (Sorvall Microspin 24S, DuPont Co.) at 10,000g for one min. Aliquots of 1 μ L each, containing 1 mg/mL, were applied in the gels. Electrophoresis conditions were 4 mA/gel, 250 V and 3 W. Protein bands were stained with Coomassie brilliant blue (Pharmacia); (Phast-system users manual, 1990). The protein gels were scanned on a 3CX Image Analyzer (Bio Image and Visage, Millipore Corporation, Ann Arbor, MI). Electrophoretic profiles and the integrated optical density (IOD) of the myosin heavy chain (MHC) and actin (Ac) bands were obtained. The molecular weights (MWs) of the main proteins in the samples was estimated by comparing their mobility with that of a standard high-MW protein mix (Ferritin, 220 kDa subunit; albumin, 67 kDa; catalase, 60 kDa subunit; lactate dehydrogenase, 36 kDa subunit and ferritin, 18,5 kDa subunit, Pharmacia). For the quantitative measurement of the MHC and Ac bands, the integrated optical density was previously checked for linearity.

Size exclusion chromatography

The 0.6M NaCl soluble fractions (S1) were analysed by size exclusion chromatography (SEC) using a Pharmacia column (length 55 cm, diam 2.5 cm) filled with Bio-Gel A-50 m gel (Medium) filler and an UV detector (Model UV-1, Pharmacia). The column was equilibrated with 0.6M NaCl pH 7.0 (Trismaleate). Pending analysis, S1 fractions were stored at -18°C in 50% glycerol (v/v) and used after overnight dialysis [0.6M NaCl pH 7.0 (Tris-maleate), 1 mM phenylmetanesulfonyl fluoride]. A 2-mg sample of dialysed protein (2-3 mL) was filtered (0.8-8 μ m) and applied onto the column, collecting fractions of 5.5 mL. The A_{280} detector's sensitivity was adjusted to 0.1 and the flowrate was held constant at 0.5 mL/min. The molecular weights of the peaks was estimated by comparing their mobility with that of dextran blue (2,000 kDa), aldolase (158 kDa) and tiroglobuline (669 kDa) (Pharmacia).

Ca²⁺-ATPase activity

Ca²⁺-ATPase activity was measured at 25°C in 1mg/ml of NAM obtained in fraction S1 according to the method of Kawashima *et al* (1973). The inorganic phosphorus (Pi) released was measured by the method of Fiske-SubbaRow (1925). Results are expressed as μ moles of inorganic phosphorous (Pi) released per minute per mg protein.

RESULTS AND DISCUSSION

Extractability

The amount of NAM extracted with 0.6M NaCl (fraction S1) (electrostatic and hydrogen bond cleavage) was initially similar in cod minces stored at -20°C and fillets stored at -20°C or -30°C (85 mg/g muscle) (Fig 1), but decreases in solubility differed with time. The decrease was greatest in minces, followed by fillets stored at -20°C then fillets stored at -30°C. Initially all the 0.6M NaCl insoluble aggregates P1 were solubilised with 2% sodium

dodecyl sulphate (SDS), which indicates that mainly secondary bonds were involved. As storage progressed there was an increase in the amount of protein extracted upon addition of 2% SDS in the minced lot (Fig 2) compared with the fillet lots (Fig 3), but a decrease in the proportion as a percentage of aggregate (P1), as in the fillet lot stored at -30°C all the protein was extracted after treating the aggregate with 2% SDS. Aggregates insoluble in 2% SDS (P2) were partially solubilised on addition of 2% SDS + 5% β -mercaptoethanol (S3), which suggests subsequent formation of disulfide bridges. A residue (P3) was left in minces and fillets stored at -20°C which increased with storage time, although it remained relatively stable as a percentage of the aggregate insoluble in 0.6M NaCl (fig 2 and 3). This residue remained even in the aggressive conditions of sample preparation for electrophoresis.

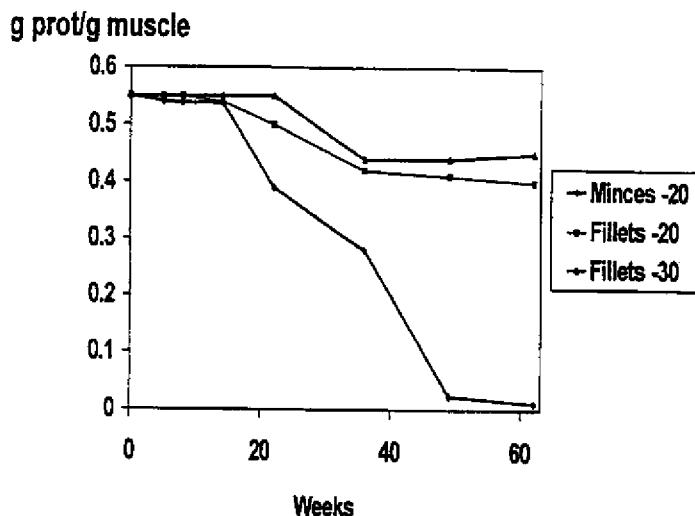


Figure 1. Grams of protein extracted with 0.6 M NaCl (S1) per gram of protein of minced cod or fillets frozen stored for 62 weeks.

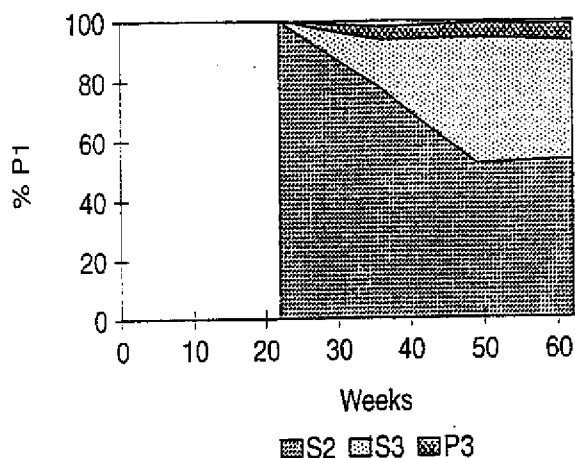


Figure 2. Protein extracted in 2% SDS (S2), 25 DSD + 5% beta-mercaptoethanol (S3) and insoluble fraction (P3) as percentages of total aggregate insoluble in 0.6M NaCl (P1). Minced cod stored at -20°C .

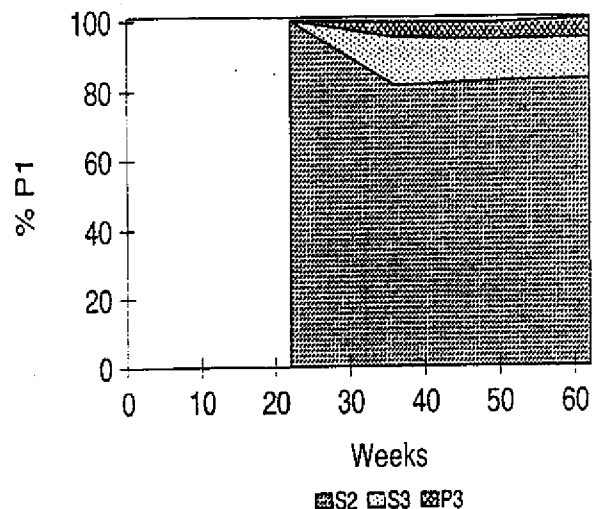


Figure 3. Protein extracted in 2% SDS (S2), 25 DSD + 5% beta-mercaptoethanol (S3) and insoluble fraction (P3) as percentages of total aggregate insoluble in 0.6M NaCl (P1). Fillets of cod stored at -20°C .

SDS-page

When the S1 fractions were analysed by SDS-PAGE, it was found that myosin heavy chain (MHC) and actin (Ac) decreased in the minces and fillets as storage time progressed, (Fig 4, 5) becoming the salt extracted protein enriched with protein of lower molecular weight than actin. This means that myosin was the protein that became insoluble fastest in frozen storage, as previously observed in isolated NAM (Del Mazo *et al*, 1994). In the S2 fraction, MHC and Ac were more stable in the minced lot than in the fillets. In minces and fillets, myosin and actin remained more stable in the S3 extracts. In all fractions, extracted proteins not entering the SDS-PAGE resolving gel were detected.

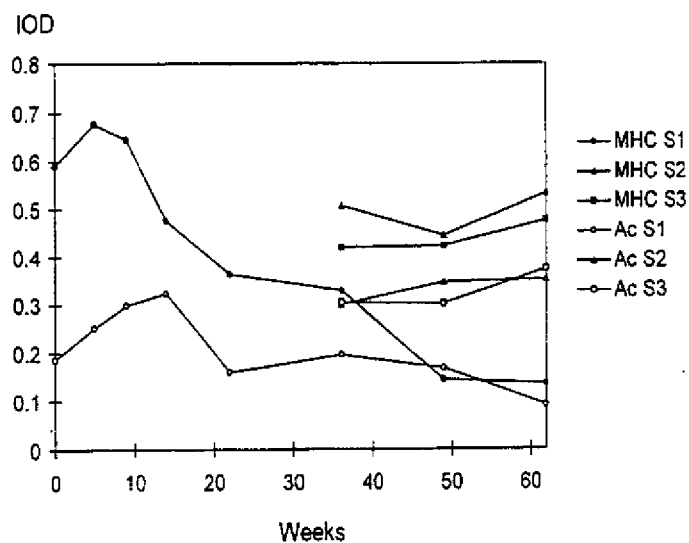


Figure 4. Integrated optical density (IOD) per microgram of protein extracted of myosin heavy chain (MHC) and actin (Ac) obtained from SDS-PAGE (12.5 %) of fractions S1, S2 and S3. Minced cod stored at -20°C .

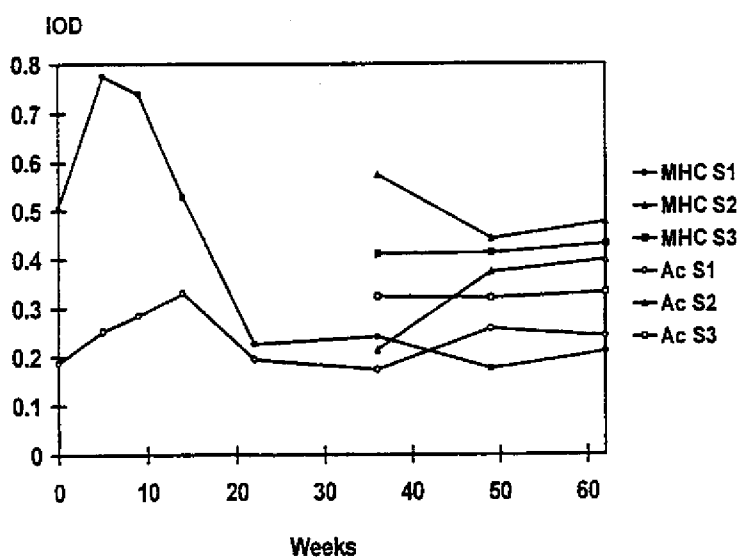


Figure 5. Integrated optical density (IOD) per microgram of protein extracted of myosin heavy chain (MHC) and actin (Ac) obtained from SDS-PAGE (12.5 %) of fractions S1, S2 and S3. Fillets stored at -20°C .

It should be remembered that although at the end of the frozen storage period there was practically no variation in the composition of fractions S2 and S3, total amount of MHC and actin varied because of the different amounts of protein extracted in the different lots (Fig 1)

Size exclusion chromatography (SEC)

Fig 6 shows the SEC chromatogram of fraction S1 for fresh cod. In frozen cod (Fig 7) initially two major peaks (peak 1 (MW>2000 kD) and peak 4(maximum MW 355 kD) were formed in fractions extracted with 0.6M NaCl (S1). Under SDS-PAGE (silver staining), peak 1 showed bands of myosin heavy chain (MHC) and actin (A) and peak 4 showed actin, tropomyosin, troponins and myosin light chains. Peak 1 diminished over time, faster in the minced lot, a development which was related with an increase in the insoluble precipitate (Table I).

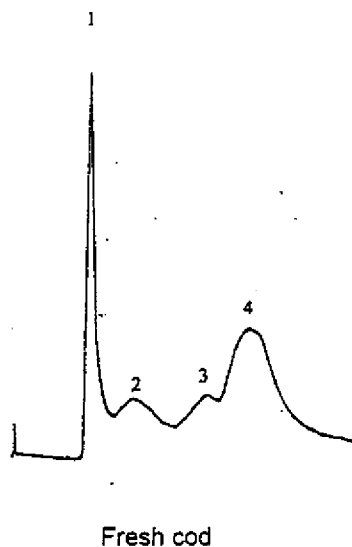


Figure 6. Size exclusion chromatograms of fraction S1 extracted from minced cod before freezing.

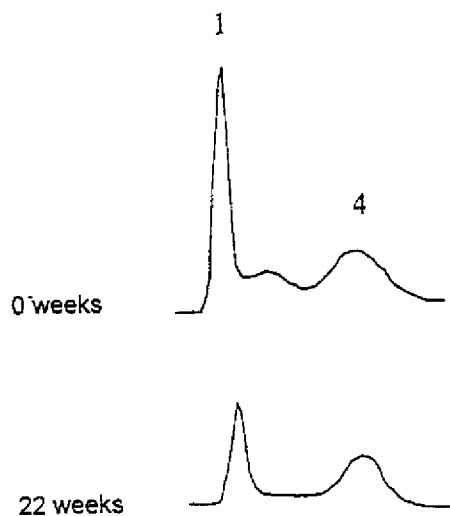


Figure 7. Size exclusion chromatograms of fraction S1, extracted at 0, 22 and 36 weeks of frozen storage. Peak 1 MW=>2000 kDa. Peak 4: maximum MW 355 KDa.

Table 1.

Lot	Weeks Frozen	Peak 1	Peak 2	Peak 3	Peak 4
Fresh Cod		24.42	10.67	14.62	50.25
Minces -20°C	0	61.60	8.90		29.46
	36	11.93			88.07
Fillets -20°C	49	57.37			42.62
Fillets -30°C	49	55.78			44.21

Ca²⁺ATPase activity

Fig 8 shows the Ca²⁺ATPase activity per mg/prot in the S1 fractions. The activity decreased in all lots with time reflecting in part the changes in composition of the extracted protein in 0.6 M NaCl. The activity per mg/prot in both lots stored at -20°C was similar and lower than in lot stored at -30°C. Nevertheless, when considering the Ca²⁺-ATPase activity per mg/muscle, the activity in the minces was much lower at the end of the storage period.

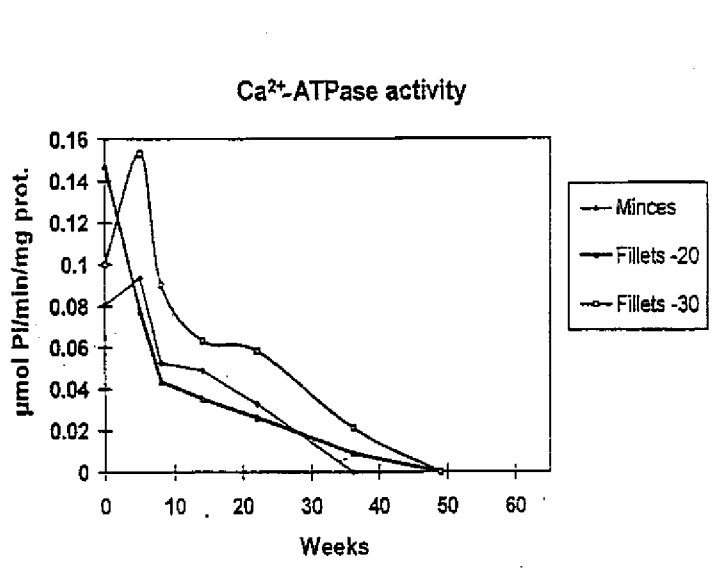


Figure 8. Ca²⁺-ATPase activity (umol Pi/min/mg prot) of cod minces and fillets frozen stored for 62 weeks

CONCLUSIONS

The results indicate that the type of bonds that form among the myofibrillar proteins of cod muscle and cause aggregation, change over time in frozen storage depending on muscle intactness and storage temperature. At the outset almost all the NAM was extracted with 0.6M NaCl. Subsequently, other secondary bonds formed, as evidenced by the need to

solubilise the protein with SDS. In the minces and fillets stored at -20°C disulfide bridges formed which were susceptible to breakdown with ME. However, in fillets stored at -30°C , all the protein was extracted when treating the aggregates with 2% SDS. The remaining insoluble residue (P3) when obtained, likely involved covalent bonds other than disulfide, as even in the aggressive conditions of sample preparation for electrophoresis, an insoluble residue still remained in P3. Both myosin and actin were the main proteins involved in aggregation although actin aggregated to a lesser extent. The decrease of Ca^{2+} ATPase activity reflects in part the changes in composition of the protein extracted in 0.6M NaCl.

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BARO- AND CRYO-PROTECTION OF PROTEINS IN PRESSURE-ASSISTED FREEZING OF SEAFOODS

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INTRODUCTION

Slow freezing of seafoods causes ice crystal formation in the extracellular fluid of seafood. To equilibrate the difference in chemical potential between the intra- and extracellular milieu resulting from such ice crystal formation, there is further migration of water from the intra- to the extra-cellular space resulting in cell shrinkage and increased hypertonicity of the intracellular solution (Rubinsky et al., 1994). Large ice crystal sizes occurring during such slow freezing coupled with the changes in osmotic pressure across cell membranes may disrupt the sarcolemma and damage seafood texture and water-holding ability.

Much smaller ice crystals are formed intracellularly by rapid freezing, but subsequent storage may also cause protein denaturation. Freeze-induced denaturation is also a problem in minced materials where ice crystal size is not important. Hence during the production of surimi, sucrose/sorbitol mixtures are added as cryoprotectants to stabilize the proteins during frozen storage. These cryoprotectants have been proposed to stabilize proteins via the "solute exclusion" or "preferential hydration" mechanism (Arakawa and Timasheff, 1982). In intact seafoods, protein denaturation is manifested as drip loss on thawing the muscle, and toughening and dryness of the texture when cooked.

To address the problem of large ice crystal formation and the concomitant denaturation of seafood proteins in intact seafoods, high pressure-assisted freezing of fish muscle is being investigated (Kalichevsky et al., 1995). In general, formation of ice crystals is associated with increased volume which, according to Le Chatelier's Principle, would be restricted under pressure. Consequently, the freezing point of pressurized water is reduced allowing products to be supercooled to very low temperatures without ice crystal formation. For example, water pressurized at 200 MPa can be supercooled to about -20°C. The first step in pressure-assisted freezing is thus pressurization of an unfrozen sample and cooling the sample under pressure to a desired sub-zero temperature. Upon subsequent release of the pressure there is rapid and uniform ice crystal formation due an increased ice nucleation rate

resulting from supercooling (about tenfold for each degree K of supercooling). Ice crystals formed under these conditions are considerably smaller than those formed by freezing under normal atmospheric pressures.

However, the pressures that may be required to supercool seafood proteins may also cause denaturation of the proteins. Solute exclusion theory would predict that common cryoprotective compounds may also baroprotect proteins. Also, the stability of muscle proteins will relate to body temperature of the animal, being greater for homeotherms and poikilotherms of higher habitat temperature. We have therefore investigated the relative effects of high pressure on the muscle proteins of beef and fish muscle from species of differing habitat temperatures, and how these effects may be influenced by various cryoprotectants.

MATERIALS AND METHODS

Muscle from Alaska pollock (*Theragra chalcogramma*), a cold-adapted fish; tilapia (*Oreochromis niloticus*), a tropical freshwater fish; and beef, were minced using a Stephan vertical cutter mixer and washed three times with 3 vols of cold water at 4°C. The washed mince was rinsed in 0.1% NaCl solution and centrifuged at 6,000 x g for 10 min. To the washed tissue were added various concentrations of sucrose, sorbitol, lactitol, maltodextrins (10 and 20 D.E), and mixtures of these, all previously demonstrated to be effective as cryoprotectants. The tissues were subjected to pressures ranging between 50 - 200 MPa for 20 min at 4°C using a special piston-driven pressurization device and the product subsequently homogenized and assayed for Ca²⁺-ATPase activity as index of protein denaturation. Protein content of homogenates was measured by the Biuret method.

RESULTS AND DISCUSSION

The effect of pressure on muscle proteins of the various species followed a similar trend as their stabilities to heat and freeze-thaw temperatures measured in previous studies (Fig. 1). That is, myosin from the cold-adapted pollock was most susceptible to pressure denaturation (loss of about 95% ATPase activity at 200 MPa) while tilapia and beef muscle proteins were more resistant to denaturation. The corresponding losses of activity for beef and Tilapia were 38% and 5%, respectively.

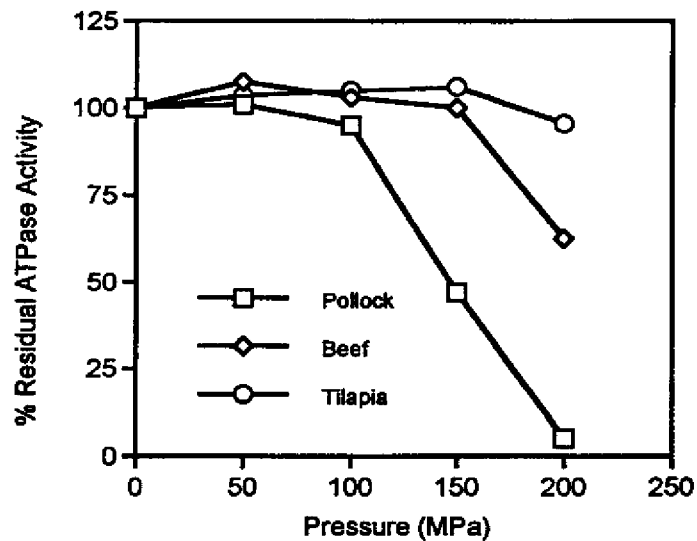


Figure 1. Effect of high pressure on fish and beef myosin.

All of the cryoprotective additives used, whether individually or as mixtures, showed some degree of baroprotection of the proteins (Fig. 2). The large molecular weight maltodextrins were the least effective while sorbitol proved to be the most effective baroprotectant. For example, in the presence of 8% sorbitol, Alaska pollock retained as much as 68% ATPase activity following pressure treatment at 200 MPa while only 29% retention of activity was observed with maltodextrins. The overall order of effectiveness of these carbohydrates as baroprotectants at 200 MPa was sorbitol > lactitol/sorbitol > lactitol = sucrose/sorbitol > sucrose > maltodextrins.

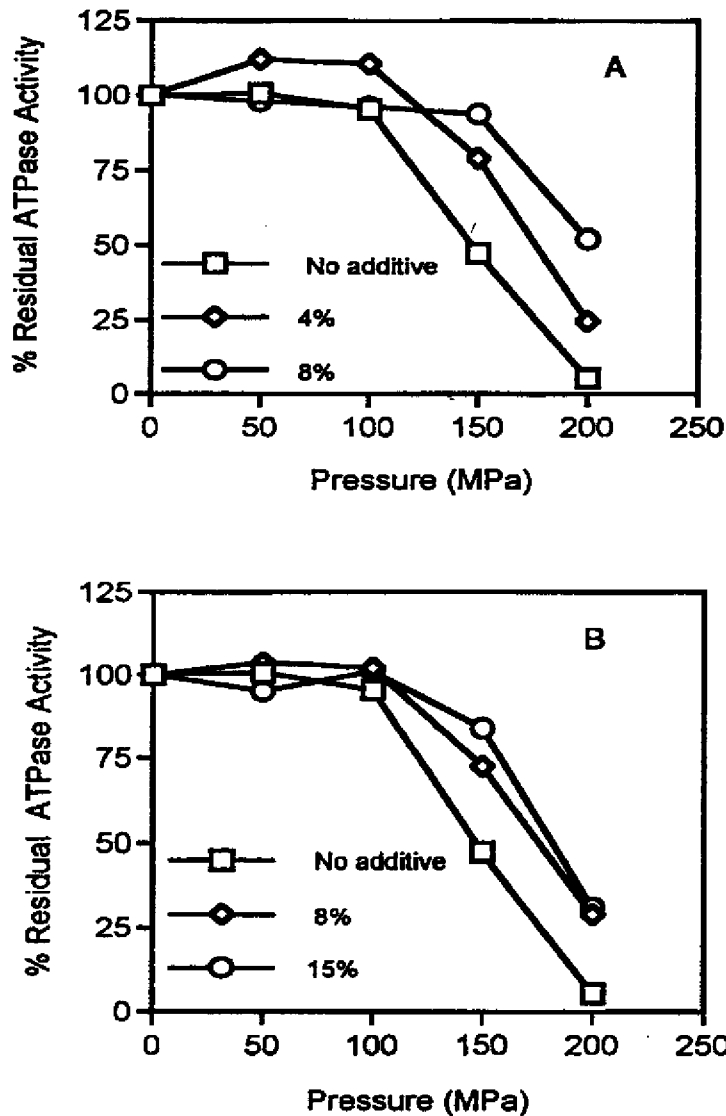


Figure 2. Effect of sorbitol and maltodextrins on high pressure denaturation of Alsaka pollock myosin. A and B represent sorbitol and maltodextrin, respectively.

Similarities between the cryoprotectant and baroprotectant effects of these compounds seem to verify the solute exclusion mechanism of protection in both cases. Furthermore, it was concluded from the results that pressure-induced protein denaturation during pressure-assisted freezing may be preventable by prior treatment of seafoods with cryoprotectants. The same additives would also confer stability during subsequent frozen storage and thawing.

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THE NATIONAL MARINE FISHERIES SERVICE'S VOLUNTARY FISH MEAL INSPECTION PROGRAM: PAST, PRESENT, AND FUTURE

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The U.S. Department of Commerce (USDC), National Marine Fisheries Service (NMFS) conducts a voluntary inspection program for the fish meal industry. This government/industry cooperative effort to control the incidence of *Salmonella* in fish meal operates under a Memorandum of Understanding (MOU) between the Food and Drug Administration (FDA), the U.S. Department of Agriculture (USDA), and NMFS. Based upon recommendations of the U.S. Animal Health Association (USAHA), the program has been recognized by a special Feed Safety Advisory Committee to the USDA as a model government/industry effort to control *Salmonella* in animal feeds. A program overview and the current status of the menhaden fish meal industry follow. The transition to a Hazard Analysis Critical Control Point (HACCP) based program and other ongoing fish meal activities of the NMFS' National Seafood Inspection Laboratory (NSIL) are discussed.

PROGRAM OVERVIEW

The National Marine Fisheries Service's Fish Meal Inspection Program operates on a reimbursable revenue basis and is available to various fish rendering facilities. A Federal or cross-licensed state inspector visits and inspects each facility three times during the normal six-month production year. These inspections consist of a sanitation evaluation in addition to collecting and analyzing fish meal samples for the presence of *Salmonella*. Upon completion of the sanitation evaluation, the inspector rates the facility as "Satisfactory," "Needs Improvement," or "Unsatisfactory." The ratings are based on compliance with the sanitation guidelines in the NOAA Publication Cir-354 entitled "Sanitation Guidelines for the Control of *Salmonella* in the Production of Fish Meal" (USDC, 1971). Ten sample units (50-200 grams each) are collected aseptically and sent to the NSIL in Pascagoula, Mississippi for *Salmonella* analysis. All sample units are analyzed using *Recommended Procedures for the Isolation of Salmonella Organisms from Animal Feeds and Feed Ingredients* (USDA, 1971). The production lot conforms to program guidelines if no more

than one of ten sample units test positive for *Salmonella*. If two or more test positive, the production lot is nonconforming. The sanitation scores are combined with the results of the *Salmonella* analyses to determine a level of compliance for the facility. The levels of compliance are "Phase I," "Phase II," or "Phase III."

COMPLIANCE LEVELS

"Phase I" plants are those just beginning the Program. While in "Phase I," the facility makes various plant or warehouse changes to meet the NMFS requirements. Once a facility has been inspected three times and *Salmonella* analytical results are available for fish meal collected on three consecutive visits, the facility moves to either "Phase II" or "Phase III" based upon the following guidelines. A plant in "Phase II" has two or more of ten sample units test positive for *Salmonella* on one of its last three inspections. A "Phase III" plant is one from which *Salmonella* is isolated from one or none of ten sample units collected on each of three consecutive plant visits. The plant must also have minimal sanitation discrepancies. Regardless of the number of *Salmonella* isolates, no plant with a current sanitation rating of "Unsatisfactory" achieves a "Phase III" status. A "Phase III" facility returns to a "Phase II" status if two or more sample units on any future inspection tests positive for *Salmonella* or if an "Unsatisfactory" rating is received during any inspection.

1994/1995 FISH MEAL INDUSTRY REPORT

During the 1994 and 1995 fish meal seasons, eight menhaden fish meal plants, representing four companies, participated in the NMFS Fish Meal Inspection Program (Table 1). While the number of facilities may seem small, they produce approximately 90% of the U.S. supply of fish meal. In addition to the eight plant locations, five off-site warehouse locations were inspected as part of the program.

Table 1. Participants

Plant Locations	Off-site Warehouses
Reedville, VA	Various Locations
Empire, LA	
Amelia, LA	
Cameron, LA	
Dulac, LA	
Abbeville, LA	
Moss Point, MS	
Reedville, VA	

The most common sanitation discrepancies observed during sanitation evaluations in 1994 and 1995 appear in Table 2.

Table 2. Most Common Plant and Off-site Warehouse Discrepancies.

Discrepancy	Number of Occurrences	
	1994	1995
Structural Integrity	29	18
General Housekeeping	12	11
Restrooms	7	10
Pest Control	24	9
Disinfectant Station	12	8
Weed Control	4	8
Doors Open	6	4
Standing Water	4	4

Levels of Compliance

Based upon NMFS inspection guidelines, including sanitation evaluation results and *Salmonella* analytical results, all plants and warehouses were rated according to their levels of compliance with the program. To maintain the confidentiality of participants, the individual plant and off-site warehouse phase characteristics are not reported. However, table 3 depicts the combined overall phase summary of plants and off-site warehouses for 1991-1995.

Table 3. Number of Facilities in "Phase II" and "Phase III" for 1991-1995
(Note: None were in "Phase I").

Year	Phase II	Phase III	Number of Participants
1991	6	4	10
1992	5	3	8
1993	4	4	8
1994	7	6	13
1995	11	2	13

Salmonella Prevalence

Thirty-nine of the 390 sample units collected from plants and off-site warehouses tested positive for *Salmonella* in 1994. This represented a combined industry prevalence rate of 10% for the 1994 season. In 1995, 86 of 390 sample units tested positive for *Salmonella* resulting in a combined industry prevalence rate of 22.1%.

FISH MEAL ACTIVITIES AT NSIL

Export Certificates

Much of the fish meal and oil produced in the United States is exported. Numerous foreign countries require an export certificate to verify adequate processing conditions to reduce the levels of *Salmonella* in fish meal. The NSIL issues export certificates to "Phase III" facilities that comply with the sanitation guidelines and have had one or less of their last thirty (30) analytical units test positive for *Salmonella*. Facilities in "Phase I or II" or non-NMFS Program participants may receive an export certificate depending upon the results of an independent lot inspection and *Salmonella* analysis of the lot of product to be exported. Export certificates for fish meal products are available in the following languages: English, Dutch/Flemish, Spanish, Italian, and French.

HACCP

The NMFS is currently converting The Fish Meal Inspection Program to a HACCP-based program. A fish meal industry workshop was conducted in 1992 to allow industry representatives to define critical sanitation issues and to identify *Salmonella* hazards at each operational step in fish meal production. From this information, a HACCP inspection model was developed and tested. Workshop and in-plant test results from the inspection model were compiled and reviewed by a HACCP Steering Committee.

The NMFS is currently developing a generic HACCP model for the production of menhaden fish meal. This model will serve as a guide to the industry in the development of individual plans specific for their operations.

As part of the 1997 Fish Meal Inspection Program, the NSIL inspectors will discuss HACCP and its concepts with company officials and will conduct HACCP-based inspections on a pilot scale. The industry's initial effort to develop and implement HACCP plans will be reviewed and assistance will be provided as needed. By the 1998 fish meal season, the NMFS' Voluntary Fish Meal Inspection Program will convert to a HACCP-based program.

Salmonella Research

During the 1993 fish meal production season, the NSIL conducted a study to compare three methods of *Salmonella* isolation in fish meal. The Food and Drug Administration's *Bacteriological Analytical Manual* (BAM) method (AOAC, 1992), the Department of

Agriculture's Recommended Procedures for the Isolation of Salmonella Organisms from Animal Feeds and Feed Ingredients (ARS) method (USDA, 1971), and a procedure recommended by the Microbiology Subcommittee of the U.S. Animal Health Association's Feed Committee were compared. In addition, the following analytical compositing systems were used: 1) three composites of five sample units; 2) five composites of five sample units; and 3) two composites of fifteen sample units. The results from this comparative study are undergoing statistical analysis and will be reported at a later date. Information gained from this and similar studies will be used to evaluate the program and to recommend changes as needed.

Video Production

The sanitation training materials used by the Fish Meal Inspection Program are being updated. A video entitled, "Sanitation in the Menhaden Fish Meal Industry" was filmed on location in fish meal processing facilities. The video emphasizes the critical sanitation items identified in the industry workshop as well as the sanitation guidelines in NOAA Cir-354 (USDC, 1971). This video will enable processors to train employees in good sanitation practices as a first step toward implementing a HACCP-based program.

A second video on HACCP for the Menhaden Fish Meal Industry is planned, addressing the critical control points (CCPs) in the processing and storage of fish meal. A possible third video will define the requirements for exporting fish meal and oil.

SUMMARY

The National Marine Fisheries Service provides assistance to the fish meal industry through its Fish Meal Inspection Program, based in Pascagoula, Mississippi. Services provided include inspections, training, sample analysis, advisory activities, issuing of export certificates, etc. The National Marine Fisheries Service is committed to maintaining, updating, and expanding these services as needed.

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DEVELOPMENT OF A SANITATION TRAINING VIDEO FOR THE FISH MEAL INDUSTRY

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INTRODUCTION

This presentation discusses the development of a sanitation training video for the fish meal processing industry by the National Seafood Inspection Laboratory (NSIL) and the potential benefits of using video training to instruct plant workers on the importance of good sanitation practices. Good plant sanitation is an essential prerequisite to any successful Hazard Analysis Critical Control Point (HACCP) program.

The video was filmed on location at various fish meal processing facilities. It highlights critical sanitation points that have been identified jointly by the fish meal industry and the National Marine Fisheries Service (NMFS). The video is designed to provide a basic understanding of Good Manufacturing Practices (GMPs) and the use of personal hygiene practices to reduce the prevalence of *Salmonella* in the finished product.

THE U.S. FISH MEAL INDUSTRY

In the United States the menhaden fish meal industry is based on *Brevoortia patronus* in the Gulf and *Brevoortia tyrannus* in the Atlantic. The fish meal industry is a valuable part of the U.S. fisheries, and menhaden landings are second in quantity only to Alaskan pollock (*Theragra chalcogramma*). Total 1995 U.S. menhaden landings were in excess of 81,000 metric tons, with a dockside value of \$99,100,000. Gulf of Mexico menhaden landings for 1995 totaled 45,360 metric tons, valued at \$51,900,000. Atlantic menhaden landings totaled 36,573 metric tons valued at \$47,200,00 (USDC, 1996).

In 1995, menhaden accounted for 98% of the total U.S. fish oil production and 68% of the total fish meal production (USDC, 1996). Menhaden products include fish oil, fish meal, solubles, bait, and animal feed. The fish oil is further processed into cosmetics, margarine

and edible oils, or used in leather tanning and industrial oils. A large percentage of fish meal, oil, and solubles are exported to Mexico, Netherlands, France, and Spain. Nineteen ninety-five fish meal and fish oil exports were valued at over \$85,450,000 (USDC, 1996).

HACCP

In an attempt to minimize the prevalence of *Salmonella* in the finished meal, individual companies are in the developmental stages of designing a fish meal HACCP-based plan. Fish meal is often used as a component of agricultural animal feeds. Fish meal contaminated with *Salmonella* can potentially pass the bacteria to consumers through the consumption of the animals. Elimination of *Salmonella* in animal feeds through the use of an employee sanitation training program in conjunction with a HACCP-based program can reduce the risk of exposing consumers to this potentially harmful bacteria.

HACCP is a preventative system of food control that requires a hazard analysis to be conducted on the product and process, and that Critical Limits (CLs) are set at each Critical Control Point (CCP). When "real time" monitoring of the critical control points reveals that critical limits are being violated, specific corrective actions are taken to isolate all noncomplying products (Garrett, 1992).

Once the product is identified as noncomplying, a specific hazard analysis is performed on the product to determine its proper disposition as well as to determine what went wrong to produce the noncomplying product. Once the cause of the process malfunction is determined, a "short-term" correction is immediately instituted and a long-term solution is identified and scheduled for implementation. All of these activities are documented through corrective action reports, including the final disposition of the noncomplying product (NMFS/NSIL, 1992; Jahncke, et al., 1996).

The seven (7) HACCP Principles are as follows:

1. Perform Systematic Hazards Analysis
2. Determine Critical Control Points
3. Establish Critical Limits
4. Determine Corrective Actions
5. Establish Monitoring Procedures
6. Establish Record Keeping System
7. Establish Verification Procedures

Good sanitation practices have always been an important key in reducing the risk of *Salmonella* contamination in the finished fish meal product. As the fish meal industry develops and implements a HACCP-based program, good sanitation practices will become even more important. To assist the industry in implementing a HACCP-based program, the NSIL is developing video tape for training fish meal employees in good sanitation practices.

In 1971, NMFS published NOAA Technical Report NMFS CIRC-354, "Sanitation Guidelines for the Control of *Salmonella* in the Production of Fish Meal"(USDC, 1971) to provide basic instruction of sanitation principles in a fish meal processing environment. At the request of several fish meal industry members, NMFS was asked to update NOAA Technical Report NMFS CIRC-354 by producing a sanitation training video for their employees to view at the beginning of each season. It was agreed that the video would focus on sanitation CCPs identified or developed during the Fish Meal HACCP Workshop held in 1992 (NMFS/NSIL, 1992).

SANITATION TRAINING VIDEO TAPE FOR THE FISH MEAL INDUSTRY

The Sanitation Training Video being developed by NMFS is based on sanitation control points which were identified by industry leaders participating in series of interactive workshops. Once the focus of the video was identified, the story board and scene list was developed by NSIL and industry personnel. Several site tours were conducted before filming the training video. These plant tours were helpful to select the appropriate filming location and to identify necessary processing details. The raw footage was taped, the narrative is finalized, and the editing process has begun.

A training video tape is an effective training tool for training plant employees. It can be widely distributed, provide consistent, concise, easily understood, and retainable instructions to personnel located in different plants. Employees who do not speak English as a first language, or do not have the time or adequate skills to read technical documents find video training tapes to be highly beneficial. A video tape can be more effective than a written instruction manual, since it does not depend on the trainer's memory or his/her interpretation of a particular task.

A successful employee training videotape requires several elements: 1) the purpose should be clear and concise, since employees are more open to training when they understand the reason behind the learning event; 2) the videotape should use familiar words, settings or situations; 3) training should be presented in a clear, easy-to-understand manner that is applicable to the employee's duties; and 4) the material should be relevant to the fish meal industry and be presented in a format that is easily retained by employees.

Upon completion, the training videotape will be distributed to various fish meal companies. Other potential audiences are USDOC inspectors, universities, and foreign fisheries personnel. The goal is to provide employees with the needed training to improve sanitation to reduce the incidence of *Salmonella* in the finished fish meal product. In addition, updated training materials specific to fish meal processing are needed to prepare for the advent of a HACCP-based Fish Meal Inspection Program.

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**AQUACULTURE POTENTIAL FOR THE FLORIDA BAY
SCALLOP, *Argopecten irradians concentricus*,
ON THE WEST COAST OF FLORIDA**

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INTRODUCTION

Scallops have a high market value and are considered a delicacy in many parts of the world where either the entire meat is consumed or only the large adductor muscle is consumed. Over 35 species of scallops are harvested world-wide but wild stocks in many areas have declined either because of over-fishing or because of natural or man-induced environmental changes. One species, the American bay scallop (*Argopecten irradians*) is a commercially important species along the east coast of the United States but commercial fishing of wild stocks of the bay scallop is presently almost non-existent with harvests in New York alone declining from 500,300 lbs. of meats in 1982 to only 300 lbs in 1988 (Wenczel, et al., 1993).

Commercial aquaculture ventures have been started in the United States in Virginia, New York, and Massachusetts but until recently economic success with the species has been less apparent because the shucked adductor muscle meats of domestically grown scallops can't economically compete with the imported American bay scallop meats from China. However, in the past five years, a market has been created in the northeastern United States and Virginia (Oesterling and DuPaul, 1993) for the whole, unshucked product like clams and oysters. This new product is marketed at a premium price as a specialty item and local demand has exceeded production, making the economics of domestic aquaculture feasible (Oesterling and DuPaul, 1994).

The commercial harvest of the species from wild stocks in Florida has been banned by the Florida Marine Fisheries Commission and no attempt has been made to examine the aquaculture potential of the species in Florida. The purpose of this study was to determine the feasibility of developing a bay scallop hatchery on a Florida estuary and to determine the time required to grow a marketable size scallop in Florida.

MATERIALS AND METHODS

Bay scallops were collected from Homosassa, Florida, on September 17 and October 25, 1991 and were brought to the laboratory in the Marine Science Department, University of South Florida at Bayboro Harbor, Tampa Bay (Figure 1). The scallops were kept in 500 liter fiberglass tanks filled with seawater (25-28‰S, 24-28°C) from Bayboro Harbor. One half of the water in each tank was replaced with fresh seawater from the same source every day. The scallops were fed with an equal volume of cultured *Isochrysis galbana* (2 million cells ml⁻¹) and *Tetraselmis sp.* (0.5 million cells ml⁻¹) at 500 ml per individual per day until they were reproductively mature. Spawning was allowed to occur when gonads became ripe and the fertilized eggs were allowed to hatch at a density of 10-20 ml⁻¹. No antibiotics were used. The bay scallop progeny (F₁) were reared initially in the lab and then in Bayboro Harbor of Tampa Bay. In the fall of 1992, mature F₁ scallops were brought into the lab from Bayboro Harbor for spawning. Larval culture, intermediate culture and growout of the F₂ generation followed the same technique used for the F₁ generation, and are described as follows:

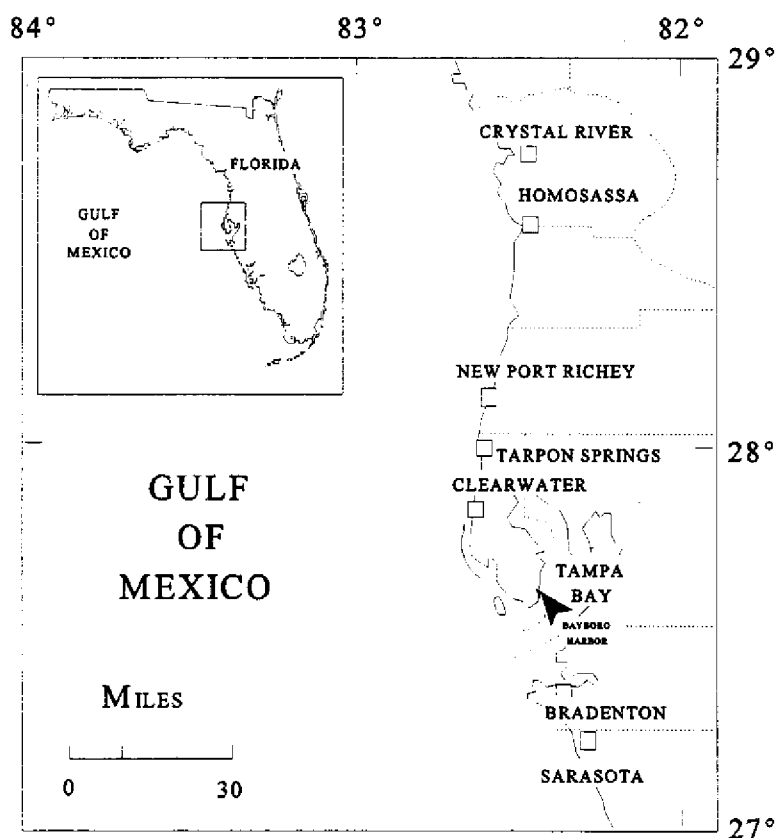


Figure 1. Location of aquaculture feasibility study.

Larval Culture

Fertilized eggs were allowed to develop for 20-30 hours when they became D-shaped larvae. They were then filtered onto a 35 µm screen and released into fresh seawater. Larvae

were maintained at a density of 4-8 ml⁻¹ and fed with 10,000-30,000 cells ml⁻¹ of *I. galbana* daily, with 1,000-5,000 cells ml⁻¹ of *Tetraselmis sp.* added as supplement. Water was replaced every day with Bayboro Harbor water in the amount of 1/3 of the total volume. Aeration was provided to the larval culture continuously. Each day a sample was taken from the culture and shell length of 20 larvae was measured (to ± 3 μ m) using a microscope fitted with a ocular micrometer. As soon as eye-spots started to develop in 8-12 days, pieces of black plastic *Thalassia* mimics or plastic outdoor carpeting (Astroturf®) were added to the culturing tanks to provide a settlement substrate for spat. Two to seven days were required for the eyed larvae to settle on the substrates and to complete metamorphosis. As soon as metamorphosis was observed, daily food ration was increased gradually from 30,000 cells ml⁻¹ to 100,000 cells ml⁻¹ of *I. galbana* and *Tetraselmis sp.*

Intermediate Culture

Once spat reached a mean shell length of >500 μ m, a process that required another 2-3 weeks, the substrate materials bearing byssate spat were placed into 300 μ m nylon mesh bags (25 cm \times 55 cm in size) at a density of approximately 10,000-30,000 spat per bag. The bags were suspended from a dock in Bayboro Harbor. In three to four weeks the 300 μ m bags were replaced with 800 μ m nylon mesh bags. Spat density was gradually reduced to approximately 1,000-2,000 spat per bag during the intermediate culture. The bags were removed from the water briefly every week and cleaned with a brush to remove sediments and fouling organisms. Every 2-3 weeks, 50 spat were sampled from a marked bag and their shell height was measured. The percent survival was also determined at that time. After the determination for growth and survival, the spat were then returned to the same bag.

Growout

When juvenile scallops achieved a mean shell height of 7-10 mm, they were transferred from bags to small mesh lantern nets (mesh opening 5 mm). The lantern nets had four levels each, with a diameter of 50 cm and height of 20 cm between levels. About 500-1,000 scallops were placed in each level, making about 2,000-4,000 scallops per net. The small mesh lantern nets were replaced with growout lantern nets with 15 mm mesh openings in 4-6 weeks. The number of scallops was gradually reduced as they grew in size, until a final density of 50 scallops per level was reached. The nets were cleaned every 2 or 3 weeks, and 50 scallops were sampled and measured in shell height with calipers to ± 0.5 mm. Empty shells were counted and removed from the nets.

RESULTS AND DISCUSSION

Growth and development of both generation (F₁ and F₂) bay scallop larvae were similar. A total of 10.4 million eggs were spawned in 1991, producing 7.5 million D-shaped larvae and 19.3 million eggs were spawned in 1992, producing 9.0 million D-shaped larvae. Mean hatching rates were 72.1% and 46.7% respectively (Table 1). D-shaped larvae developed in

20-30 hours and eyed-larvae developed in 8-12 days after fertilization. Mean larval growth rate was 8.0-8.8 $\mu\text{m d}^{-1}$.

Table 1. Survival during early developmental stages of bay scallops grown in Bayboro Harbor of Tampa Bay, Florida

Developmental Stages	F ₁ generation (1991-92)		F ₂ generation (1992-93)	
	Survival (%)	Cumulative survival (%)	Survival (%)	Cumulative survival (%)
Eggs - D-larvae	72.1	72.1	46.7	46.7
D-larvae-Eyed larvae	91.0	65.6	87.8	41.0
Eyed larvae-0.5 mm spat	33.1	21.7	59.5	24.4
Intermediate culture (0.5 - 10 mm, in bags)	22.6	4.9	17.8	4.3

Metamorphosis (175-200 μm) occurred 12-13 days after fertilization. A slightly faster growth of F₂ larvae can be attributed to the 2-3°C higher temperature in 1993 than in the previous year. Survival of larvae over metamorphosis ranged from 35-60%. Overall survival from eggs to juvenile scallop of 8-10 mm shell height was less than 5% (4.9% and 4.3% in F₁ and F₂ scallops respectively).

Castagna and Duggan (1971) found that larvae of the Virginia and North Carolina bay scallop began settling in 10-19 days with most occurring in 10-14 days at 20-28°C. They also found that from early post-setting period to 2 mm in height, mortalities often reduced the number of live scallops by an estimated 50-80%. Despite the different food species and ration used in the two studies, the rate of development and survival during the early stages appear similar for the Florida and Virginia populations.

Spat were grown in the hatchery until they achieved a mean shell height of 0.8-0.9 mm after which they were placed in the bay in nylon mesh bags. Mean growth rate from pediveliger to this stage was 34.7-35.9 $\mu\text{m d}^{-1}$. Apart from the two best known critical periods limiting early survival, i.e., hatching of eggs and metamorphosis of larvae, intermediate culture in mesh bags often represents another period of heavy spat loss. Survival in intermediate culture was around 20% in both generations that were examined. Intermediate culture is often one of the key factors limiting production of scallop spat in commercial hatcheries. High mortality could be the result of crowding and low food flux and oxygen levels associated with reduced circulation due to fouling of the bags.

Scallop growth during the intermediate culture in mesh bags and during growout in lantern nets is given in Figure 2. The growth pattern of the bay scallops during growout is similar to that of a natural population in Anclote Estuary on the Gulf coast of Florida (Barber

and Blake, 1983). In both studies, growth from January to March was slow, then became rapid in April in accordance with the increase of water temperature, and high growth persisted until June and July. After that, growth declined again. In this study, growth rate from March to May, corresponding to scallop height of 17-40 mm, was 0.29 mm d^{-1} for F_1 scallops and 0.33 mm d^{-1} for F_2 scallops, while over the same period in the Anclote Estuary growth rate of wild bay scallops was about $320 \mu\text{m d}^{-1}$ (estimated from the data of Barber and Blake, 1983). In Tampa Bay, scallops grew from 9.5 mm (F_1 , 91-92) or 8.5 mm (F_2 , 92-93) to 50 mm in about 7 months, a growth rate faster than those reported for Georgia where scallops grew from 9.8 mm to 49 mm in 8 months (Heffernan *et al.*, 1988) and for North Carolina where scallops grew from 9 mm to 50 mm in 8 months (Gutsell, 1930).

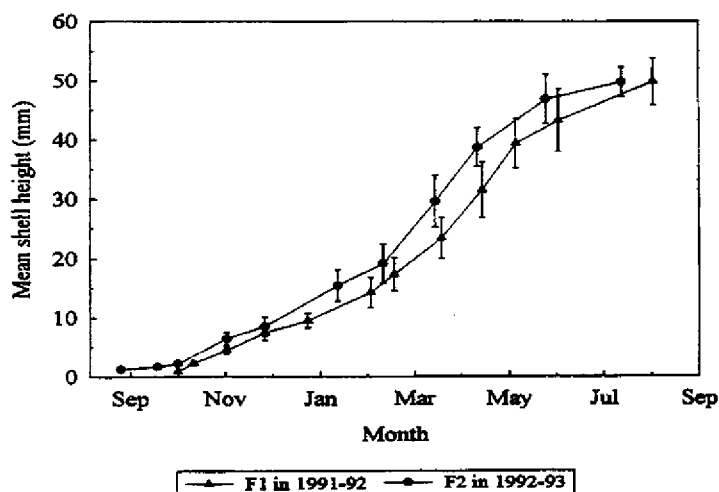


Figure 2. *Argopecten irradians concentricus*. Growth of F_1 and F_2 scallops in Bayboro Harbor in 1991-92 and 1992-93.

Mortality of scallops transferred to lantern nets is shown in Figure 3. Mortality was initially low but steadily increased through the spring. Cumulative mortality for both generations from March through September ranged from 88-90%.

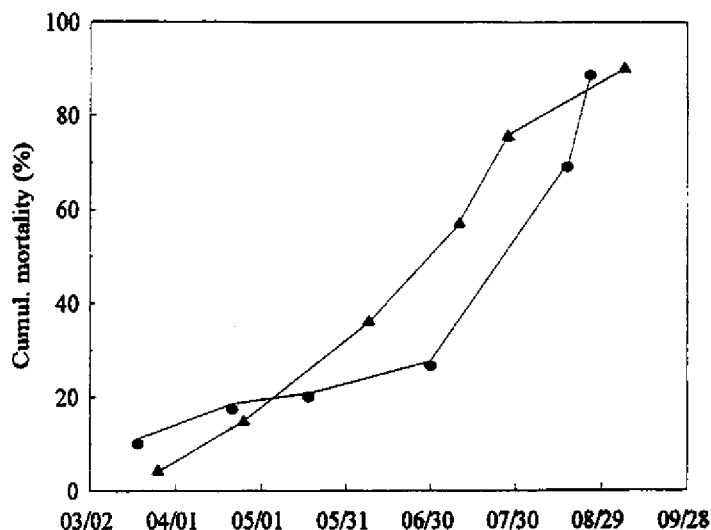


Figure 3. *Argopecten irradians concentricus*. Cumulative mortality of F₁ and F₂ scallops in Bayboro Harbor, Tampa Bay in 1992 and 1993.

Both F₁ and F₂ scallops cultured in Tampa Bay reached a maximum mean shell height of about 50 mm, with an individual maximum of 58 mm recorded. Fouling suspension feeders reduce water circulation and compete for food resources with scallops (e. g. Belding, 1910; Broom, 1976), and this may be one of the main causes that prevent scallops from growing to a comparable maximum mean size of >60 mm as observed in the Anclote Estuary population (Barber and Blake, 1983). In culturing bay scallops in coastal waters of Georgia, Heffernan *et al.* (1988) suggested that the lower growth and survival of bay scallops reared in "bottom" pearl nets may be related to varying interference from fouling organisms.

Fouling represents a serious problem in using caged scallops for growout, especially in an estuary such as Tampa Bay (Table 2). Main fouling organisms found on scallop shells were barnacles, oyster spat, tunicates and polychaete worms. The most extensive fouling starts in May and extends throughout the whole summer. A scallop net of about 10 pounds may be 30-50 pounds in just one month, mainly due to the growth of tunicates on both scallops and nets. Scallop nets had to be cleaned or replaced every 3-4 weeks during the summer.

Table 2. Major fouling organisms found on shells of bay scallops grown in Bayboro Harbor, Tampa Bay in August, 1993

Scallop	Barnacle	Polychaete	Oyster	Tunicate	Total
#1	34	2	18	4	58
#2	56	11	20	2	89
#3	38	4	14	5	61
#4	31	5	25	1	62
#5	52	1	19	3	75
#6	34	4	24	3	65
#7	45	2	12	4	63

Fouling organisms affect the scallops in several other ways than just competing for space and food. Tunicates grow very fast. It was often seen that one tunicate can bind several scallops together, and some scallops were smothered. Oysters do the same; they outgrow scallops and often seal the scallop valves, thus either smothering the scallops or making it impossible for them to close their shells. Polychaete worms are common borers of scallop shells (Blake and Evans, 1973). They penetrate shells and produce black blisters in the inner shells of the scallops. In this study, one hundred percent of the live scallops were bearing such worms and blisters in later summer, coincident with the period of high mortality of the scallops. It seems that the heavy infection of polychaete worms and the blisters they produced contributed to the high mortality observed. The degree of fouling and possible fouling reduction techniques must be a primary consideration for any scallop aquaculture venture involving caged scallops in an estuary such as Tampa Bay.

Polychaete worm (*Polydora*) infection was also reported for the bay scallop in Massachusetts (Turner and Hanks, 1959), in Rhode Island (Russell, 1973) and in Long Island, New York (Tettelbach and Wenczel, 1993), and in the Japanese scallop *Patinopecten yessoensis* in Japan (Imai, 1971). Russell (1973) found no evidence to link the high mortality of Rhode Island scallops to the worm infection. However, Turner and Hanks (1959) and Tettelbach and Wenczel (1993) found worm infection did cause the death of bay scallops. In the case of Long Island, infection was noticed in January, 1991 and by March 100% of the 1773 scallops sampled were infected. Tettelbach and Wenczel (1993) also found holes 1-2 cm in diameter in the middle of the dorsal valve at or near where the adductor muscle attaches to the shell. They suggested that the holes resulted from the forceful contracting force of the adductor muscle, since worm infections lead to weakened scallop shells (Bergnan *et al.*, 1982).

CONCLUSIONS

Results presented in this study demonstrate that the bay scallop can survive, grow and reproduce in a Florida estuary such as Tampa Bay. Scallops can be grown to a marketable size (as summarized in Figure 4) in about 6-7 months. However, heavy mortality probably as a result of severe fouling limits the yield. Thus Tampa Bay and especially Bayboro Harbor is not an ideal location for commercial growout. In addition, if the scallops are to be used as a specialty item in restaurants serving the whole shell product, fouling of the shells will limit the appeal. Other locations such as the Homosassa/Crystal River area should be considered as possible areas for growout to produce a supply for a marketing study.

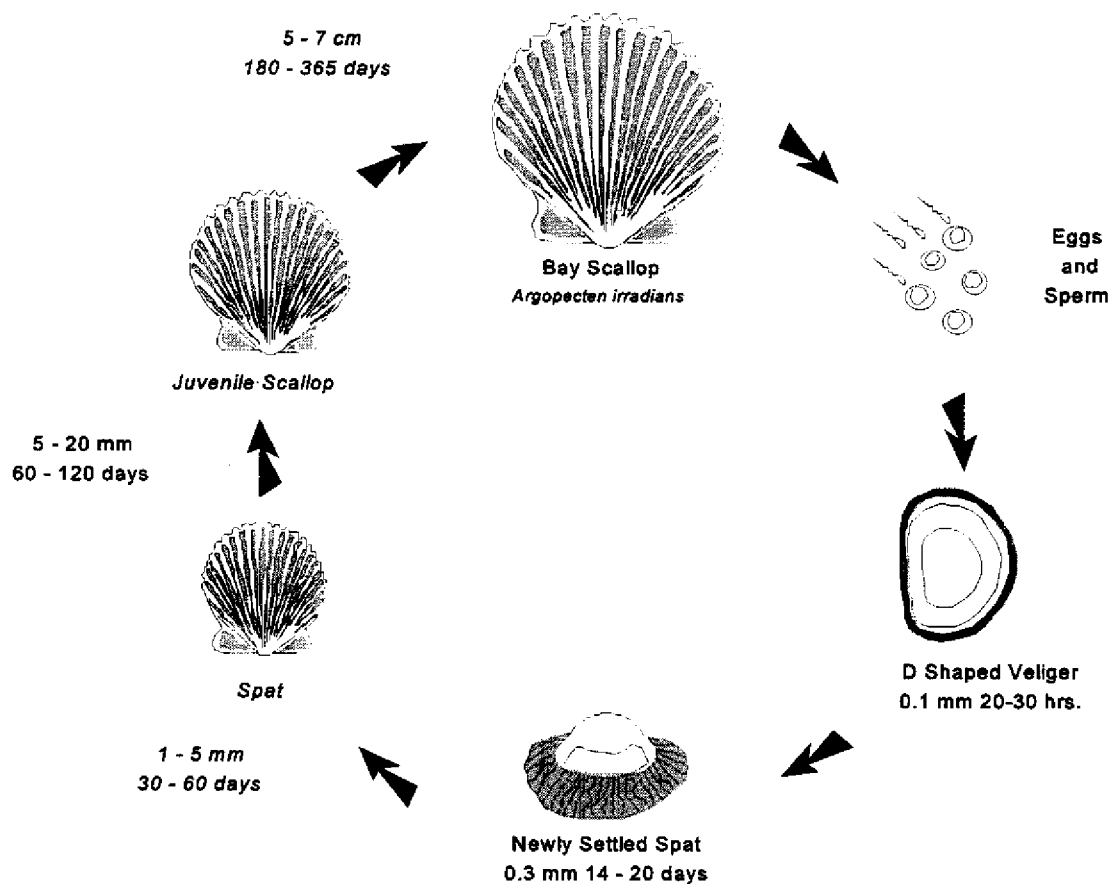


Figure 4. Life cycle of scallops aquacultured to marketable size on the west coast of Florida.

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**THE SOUTHEAST U.S. SHRIMP PROCESSING SECTOR: AN
ECONOMIC ANALYSIS OF STRUCTURE
AND CHANGES THEREIN**

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Shrimp is the predominant fishery in the Southeast United States, i.e., the coastal states extending from North Carolina through Texas, generally accounting for more than 50% of the region's landings by value. Southeast shrimp landings in 1993 equaled 175 million pounds (headless weight) and had an associated dockside value of \$454 million.

Though Southeast shrimp landings are large and production also occurs to a lesser extent in the Northeast and Pacific Regions of the United States, the U.S. depends largely on imported shrimp to meet its domestic market needs. Since the mid-1980's, in fact, more than 80% of the total U.S. shrimp supply has been import based. These imports, which originate primarily from two regions of the world, Latin America and Asia, have expanded rapidly since the late 1970's in response to increased world shrimp production and various economic factors (2).

Domestic landings and imports, when combined, support a large shrimp processing industry in the Southeast United States, with 1993 output valued in excess of \$1.0 billion. The overall goal of this paper is to examine the economic structure of the Southeast shrimp processing sector and changes that have occurred in relation to the growth in imports. To accomplish this goal, a brief overview of shrimp landings and imports is presented in the next section of the paper. Then, the shrimp processing industry is analyzed within an historical context based on National Marine Fisheries Service annual end-of-the-year survey of seafood processing and wholesaling establishments. In the final section of the paper, a more detailed analysis of 1991 Southeast shrimp processing activities is presented based on primary data collected and analyzed by the authors.

HISTORICAL SOUTHEAST U.S. SHRIMP LANDINGS AND IMPORTS

Reported Southeast commercial shrimp landings for 1973-93 are presented in Table 1 in three-year intervals. As indicated, U.S. warm water shrimp production (i.e., Southeast landings), exhibited little or no upward trend during the time frame considered with average annual production ranging from a low of 129 million pounds during 1973-75 to a high of 188 million pounds in 1985-87. While the long-run Southeast U.S. shrimp poundage has been relatively constant, the associated deflated value has fallen sharply due to a decline in the deflated per pound price (Table 1). The per pound deflated price of \$2.59 in 1991-93 was less than 70% of the \$3.89 reported in 1973-75 and only 60% of the peak deflated price of \$4.25 per pound reported in 1976-78.

Table 1. Reported Southeast Commercial Shrimp Landings, 1973-93 (3 yr. avgs.).

Time Period	Pounds Landed ^a	Value		Dockside Price	
		Current	Deflated ^b	Current	Deflated
	Mills	----- \$ Mill -----	-----	----- \$/lb -----	-----
1973-75	128.9	187.9	501.5	1.46	3.89
1976-78	165.3	327.4	702.3	1.98	4.25
1979-81	160.4	412.1	664.0	2.57	4.14
1982-84	153.9	482.0	630.9	3.13	4.10
1985-87	188.3	530.4	628.4	2.82	3.34
1988-90	166.7	449.2	473.2	2.70	2.84
1991-93	156.5	434.5	405.8	2.78	2.59

^a Expressed on a headless weight basis.

^b The deflated value and price are expressed in 1990 dollars, based on the Consumer Price Index.

Source: (4)

The decline in the deflated Southeast shrimp price and associated value is in response to sharply rising imports (Table 2). As indicated, 1991-93 imports averaging 679 million pounds annually (expressed on a headless shell-on equivalent weight basis) exceeded 1973-75 average annual imports of 243 million pounds by about 140%. Furthermore, the vast majority of the increase was after the 1979-81 period and reflects the growth in the Asian farm-raised shrimp production and subsequent export of the product to the U.S. market (2, for a more detailed discussion of the shrimp import market). In general, the domestic and imported shrimp prices average within 5%-10% of one another when examined in three-year intervals, suggesting one to be a close substitute for the other. This would imply that

imported product could be used in processing activities when domestic supply is limited, or vice versa.

Table 2. U.S. Shrimp Imports, 1973-93 (3 yr. avgs).

Time Period	Value			Price	
	Quantity ^a	Current	Deflated ^b	Current	Deflated
	Mill. lbs	----- \$ Mill. -----	-----	----- \$/lb -----	-----
1973-75	243.3	338.4	897.7	1.39	3.69
1976-78	261.4	458.8	989.0	1.76	3.78
1979-81	262.1	718.8	1,115.5	2.74	4.41
1982-84	387.7	1,143.9	1,493.1	2.95	3.85
1985-87	509.1	1,442.5	1,708.8	2.83	3.36
1988-90	580.4	1,706.2	1,798.1	2.94	3.10
1991-93	678.6	2,015	1,874.5	2.97	2.76

^a Expressed on a headless shell-on weight basis.

^b The deflated value and price are expressed in 1990 dollars, based on the Consumer Price Index.

Source: (4)

U.S. imports of shrimp by product form, expressed on a product weight basis, are presented in Table 3. Shell-on imports advanced from an average of 124 million pounds annually in 1973-75 to 335 million pounds in 1991-93. Imports of peeled shrimp advanced from 82 million pounds to 233 million pounds with the most recent three year period of analysis exhibiting particularly strong growth. Both of these product forms can be used in a variety of processing activities. Breaded and canned imports are relatively small in proportion to the total and, because they are fully processed before entering the U.S., are not subject to any additional processing by U.S. processors.

Table 3. U.S. Shrimp Imports by Product Form, 1973-93 (3 yr. avgs.)

Time Period	Product Weight			
	Shell-on	Peeled	Breaded	Canned
	----- Mill. lbs -----			
1973-75	124.2	82.4	1.6	3.4
1976-78	118.9	96.4	1.3	2.6
1979-81	134.4	82.3	3.3	4.3
1982-84	209.2	97.1	8.8	10.7
1985-87	268.3	127.2	1.8	16.7
1988-90	352.8	137.5	1.3	11.7
1991-93	335.5	232.8	1.4	8.9

Source: (4)

HISTORICAL SOUTHEAST U.S. SHRIMP PROCESSING ACTIVITIES

Shrimp represents the primary component of the Southeast seafood processing industry, generally contributing more than 80% of the total edible production activities by value. An economic analysis of the shrimp processing industry is presented below in three sections. In the first section, some general characteristics of the industry are presented. Then, some features regarding the structure, conduct, and performance of the Southeast shrimp processing industry are outlined. The analysis presented herein is based on annual National Marine Fisheries Service end-of-the-year surveys of seafood processing plants and covers the 1973-93 period.

Historical Industry Characteristics

The number of firms engaged in Southeast shrimp processing activities, as indicated by the information in Table 4, declined during the 1973-93 period. Overall, the 124 firms processing shrimp, on average, during 1991-93 reflected a 24% decline from the 164 reported during 1982-84 and about a 30% decline from the 175 reported annually during 1973-75.¹ A decline in the number of firms was evident during each interval of analysis except during 1979-81 when a marginal increase of one firm was reported.

Table 4. Shrimp Processing Activities in the Southeast United States, 1973-90 (3-yr. avgs.).

Time Period	No. of Firms	Processed Quantity		Processed Value ^a		Processed Price ^b	
		Product Weight	Headless Shell-on Weight	Current	Deflated	Current	Deflated
		----- 1,000 lbs -----		----- \$1,000s -----		----- \$/lb -----	
1973-75	175	189,768	178,713	350,219	941,167	1.85	4.96
1976-78	169	229,759	226,357	611,535	1,307,790	2.66	5.69
1979-81	170	215,163	216,042	769,528	1,238,691	3.58	5.76
1982-84	164	231,727	230,331	943,291	1,233,409	4.07	5.32
1985-87	149	270,553	267,235	1,018,748	1,207,085	3.77	4.46
1988-90	148	291,041	283,644	1,011,923	1,063,406	3.48	3.65
1991-93 ^a	124	266,316	257,123	898,502	839,162	3.37	3.15

^a The 1990 Consumer Price Index was used to deflate value and price.

^b Expressed on a product-weight basis.

Source: Compiled from unpublished data provided by the National Marine Fisheries Service, Fisheries Statistics Division.

The quantity processed, as reported in Table 4, is given on both a product-weight basis and a headless shell-on basis (3, for a list of all conversion factors).² The processed quantity, expressed on either a product weight or a headless shell-on weight basis tended to increase during the period of analysis with the exception of an almost 10% decline during the most recent three-year period of analysis and a more moderate decline of approximately 5% during 1979-81. Overall, 1991-93 average annual production of 266 million pounds (product weight basis) represented an increase of 40% when compared to 1973-75 average annual processing activities of 190 million pounds. When evaluated on a headless shell-on weight basis, the increase was nearly 45%, suggesting a relative increase in processing activities that use little additional ingredients or shell, such as peeling.

The current value of Southeast shrimp processing activities increased from an annual average of \$350 million annually in 1973-75 to more than \$1 billion annually by the late 1980's before falling to about \$900 million in 1991-93 (Table 4). This increase can be explained by two factors. First, as noted, the processed quantity increased by about 40% during the 21-year period of analysis. Second, the current price of the processed product increased throughout much of the period of analysis before declining after 1982-84.

When adjusted for inflation, the value of Southeast shrimp processing activities declined steadily after the 1976-78 period despite a general increase in quantity processed. This decline reflects the sharp fall in the real price of the processed product since 1979-81. The observed deflated per pound processed price of \$3.15 (product weight basis) in 1991-93

reflected more than a 45% decline from the 1979-81 price of \$5.75 per pound and about a 35% decline when compared to the per pound deflated price of \$4.96 in 1973-75. This decline likely reflects three primary factors: (1) increased domestic production of processed shrimp, (2) competition from imported processed shrimp, and (3) declining input costs of the raw, unprocessed product.

Historical Economic Structure, Conduct, and Performance

Several aspects of economic structure, conduct, and performance of the Southeast shrimp processing industry are presented below. They include (i) productivity within the industry and changes therein, (ii) size distribution of firms within the industry and changes therein, (iii) specialization among firms and changes therein, and (iv) industry concentration and changes.

Productivity in the Shrimp Processing Industry

Productivity within the Southeast shrimp processing industry was evaluated using two methods. The first was the estimation of production per firm. The second was the estimation of production per worker.

Production per Firm. Production per firm among Southeast U.S. shrimp processors is given in Table 5 for the 1973-93 period. Shrimp processing activities per firm, evaluated on the basis of processed quantity, clearly increased during the period of analysis when examined in three-year intervals. In 1973-75, for example, the average was 1.1 million pounds annually. By 1991-93, the annual average had approximately doubled to 2.1 million pounds. An increase in per firm poundage was particularly evident in 1985-87.

Table 5. Per Firm Production of Processed Shrimp in the Southeast United States, 1973-90 (3-yr. avgs.).

Time Period	Processed Pounds ^a	Value	
		Current	Deflated ^b
	1,000s	-----	\$1,000s -----
1973-75	1,086	2,005	5,388
1976-78	1,357	3,611	7,723
1979-81	1,268	4,536	7,301
1982-84	1,410	5,740	7,506
1985-87	1,812	6,822	8,083
1988-90	1,962	6,822	7,169
1991-93	2,148	7,246	6,767

^a Given on a product weight basis.

^b The deflated value is expressed in 1990 dollars and is based on the Consumer Price Index. Source: Compiled from unpublished data provided by the National Marine Fisheries Service, Fisheries Statistics Division.

The current value of shrimp processing activities per firm grew from an annual average of \$2.0 million in 1973-75 to \$7.2 million in 1991-93. When examined on a deflated basis, however, the value of shrimp processing activities per firm changed little after 1976-78 despite the increase poundage processed. This reflects the declining deflated per pound price received for the finished products.

Production per Worker. Selected statistics pertaining to per worker productivity in the Southeast shrimp processing industry are presented in Table 6.³ As indicated, Southeast shrimp processing establishments typically employed from 40 to 50 production workers, on average, on an annual basis. Pounds of processed shrimp produced per worker increased from an average of 28.5 thousand annually in 1973-75 to almost 43 thousand in 1991-93. Examination of the data shows two distinct periods of increasing production per worker. During the first of these two periods, i.e., 1976-78, production per worker increased to 37.2 thousand pounds from 28.5 thousand pounds annually in 1973-75. This increase may have been an artifact of the relatively low domestic supply availability in 1973-75 (see Table 1) which may have resulted in an abnormally low production per worker in that interval. The second period of increasing production was post 1982-84. The increase in production per worker during this period likely reflects response to the sharp rise in U.S. imports of shell-on and peeled shrimp which provided U.S. processors an additional supply of raw material product (see Table 3).

Table 6. Estimated Per Worker Productivity in the Southeast U.S. Shrimp Processing Industry, 1973-90 (3-yr avgs.).^a

Time Period	Avg. No. Workers Per Firm	Processed Quantity ^b	Value	
			Current	Deflated ^c
		lbs	----- \$ -----	
1973-75	46	28,524	52,954	142,736
1976-78	44	37,153	100,684	215,223
1979-81	41	37,188	134,978	216,766
1982-84	44	37,542	155,393	202,930
1985-87	53	40,458	152,167	180,242
1988-90	46	42,930	153,464	161,341

^a The analysis of per worker productivity was based only on that segment of shrimp processing establishments primarily engaged in shrimp processing activities, i.e., defined as $\geq 95\%$ of the annual value of a firm's processed seafood sales were shrimp based.

^b Expressed on a product-weight basis.

^c Expressed in 1990 dollars based on the Consumer Price Index.

The per worker deflated value of shrimp processing activities in the Southeast increased from an average of \$143 thousand annually in 1973-75 to \$217 thousand in 1979-81. After 1979-81, however, the per worker deflated value of shrimp processing activities in the Southeast fell sharply, averaging just \$161 thousand in 1991-93. This decline reflects the sharp fall in the per pound price of the processed product (see Table 4).

Size Distribution of Firms

To examine the size distribution of shrimp processing firms in the Southeast and changes therein over time, firms were grouped into four categories based upon their deflated value of processed shrimp sales: (i) firms with annual deflated processed shrimp sales of < \$250 thousand, (ii) firms with annual deflated processed shrimp sales from \$250 thousand to \$1.0 million, (iii) firms with processed shrimp sales of \$1.0 million to \$10.0 million, and (iv) firms with annual deflated processed shrimp sales of \$10.0 million or more.

As indicated by the information in Table 7, approximately one-third of the Southeast shrimp processing firms generally reported annual deflated processed shrimp sales of less than \$250 thousand, when examined in three-year intervals. Another 10% to 20% reported annual deflated processed shrimp sales in the \$250 thousand to \$1.0 million range. From about 30% to 40% of the firms reported annual deflated sales in the \$1.0 to \$10.0 million range. Finally, the remaining 15% to 20% of the total number of shrimp processors reported

processed shrimp sales of \$10.0 million or more. The information contained in Table 7 suggests that, in general, there was little change in the distribution of Southeast shrimp processing firms during 1973-93 when examined in three-year intervals.

Table 7. Size Distribution Among Firms in the Southeast Shrimp Processing Industry, (includes only processed shrimp sales), 1973-90 (3-yr. avgs).^a

Time Period	<u>< \$250,000</u>		<u>\$250,000- \$1 Million</u>		<u>\$1 Million- \$10 Million</u>		<u>≥ \$10 Million</u>	
	No. of Firms	% of Total	No. of Firms	% of Total	No. of Firms	% of Total	No. of Firms	% of Total
1973-75	56	32	33	19	61	35	26	15
1976-78	54	32	24	14	55	32	37	22
1979-81	57	34	25	15	56	33	31	18
1982-84	57	35	18	11	58	35	32	19
1985-87	41	28	19	13	59	39	31	21
1988-90	42	28	22	15	53	36	32	21
1991-93	35	28	18	15	47	38	24	19

^a Evaluated on the basis of the deflated value of processed shrimp sales.

Source: Compiled from unpublished data provided by the National Marine Fisheries Service, Fisheries Statistics Division

Specialization in the Southeast Shrimp Processing Industry

To examine specialization of firms in the Southeast shrimp processing industry, firms were partitioned based upon the percentage of total processed seafood sales (by value) that were derived from shrimp processing activities. The partitioning was as follows: (i) firms that derived less than 50% of total annual processed seafood sales, by value, from shrimp processing activities, (ii) firms that derived from 50% to 95% of annual processed seafood sales from shrimp processing activities, and (iii) firms that derived 95% or more of processed seafood sales from shrimp processing activities.

As the information in Table 8 indicates, about two-thirds of the Southeast shrimp processors have traditionally relied upon shrimp processing activities for 95% or more of their total processed seafood sales, expressed on a value basis. The percentage in recent years, however, has risen to more than 70%. Another 15% to 20% of the shrimp processors have traditionally relied upon shrimp processing activities for 50% to 95% of their total processed seafood sales, with the low end of the range occurring in more recent intervals of analysis. Finally, from slightly less than 15% to about 20% of the Southeast shrimp processors have traditionally relied upon shrimp processing activities for less than 50% of their total

processed sales. The proportion of total firms in this group also fell sharply after the mid-1980's. In general, the information indicates that increased specialization in the Southeast shrimp processing industry occurred after the mid-1980's.

Table 8. Specialization Among Southeast Shrimp Processing Firms: Processed Shrimp Sales as a Percentage of Total Processed Seafood Sales, 1973-90 (3-yr. avgs.).^a

Time Period	< 50% of Sales		50% to 95% of Sales		≥ 95% of Sales	
	No. of Firms	% of Total	No. of Firms	% of Total	No. of Firms	% of Total
1973-75	36	21	31	18	107	61
1976-78	32	19	31	19	106	63
1979-81	33	19	29	17	108	64
1982-84	33	20	27	17	104	63
1985-87	26	17	21	14	103	69
1988-90	20	13	22	15	106	72
1991-93	14	11	21	17	90	72

^a Processed sales were evaluated on the basis of value.

Source: Compiled from unpublished data provided by the National Marine Fisheries Service, Fisheries Statistics Division.

Industry Concentration

Concentration among firms in the Southeast shrimp processing industry during 1973-93 and changes therein can be evaluated using several alternative methods. One such method commonly employed in applied research is the market share approach in which the market shares accumulated by the largest x firms are analyzed ($x \in N$; and N is the total number of firms comprising the industry). Starting with the firm with the largest market share, measured in value of processed shrimp sales, and adding the shares of the next largest firms in succession, produces an estimate of accumulated market shares (measured in value) in the Southeast shrimp processing industry. These shares, evaluated in terms of the largest five, ten, and twenty establishments for the 1973-93 period are given in Table 9.

Table 9. Market Shares of Largest Shrimp Processing Firms in the Southeast U.S., Ranked by Value of Processed Shrimp Sales, 1973-90 (3-yr. avgs.).

Time Period	Largest Five Firms	Largest Ten Firms	Largest Twenty Firms
----- % -----			

1973-75	30	46	65
1976-78	28	45	65
1979-81	30	47	67
1982-84	30	47	66
1985-87	29	47	66
1988-90	28	44	63
1991-93	32	50	69

Source: Compiled from unpublished data provided by the National Marine Fisheries Service, Fisheries Statistics Division.

The largest five Southeast U.S. shrimp processing firms consistently represented from 28% to 32% of the Southeast U.S. shrimp processing activities when measured in terms of value while the largest ten firms represented from 45% to 50% of the total. The largest twenty firms consistently represented about two-thirds of the total. Overall, the market share approach indicated that concentration in the Southeast shrimp processing industry has remained relatively stable during the period of analysis.

Historical Processing Activities by Product Form

For purposes of evaluating historical NMFS data, shrimp processing activities were examined on the basis of four product forms: (i) raw headless products, (ii) peeled products, (iii) breaded products, and (iv) specialty products. Selected statistics related to the Southeast processing of these products are contained in Table 10.

Table 10. Shrimp Processing Activities in the Southeast United States,
by Product Form, 1973-90 (3-yr. avgs.).

Time Period	No. of Firms ^a	Processed Quantity		Processed Value	
		Product Weight	Headless Shell-on Weight	Current	Deflated ^b
		----- 1,000 lbs -----		----- \$1,000s -----	
		----- Raw Headless -----			
1973-75	106	66,850	66,850	134,025	359,484
1976-78	108	103,610	103,610	311,204	664,573
1979-81	118	89,457	89,457	352,943	568,845
1982-84	126	92,656	92,656	412,950	539,753
1985-87	113	105,481	105,481	435,438	516,542
1988-90	111	100,563	100,563	428,930	451,926
1991-93	85	76,161	76,161	329,811	308,730
		----- Peeled -----			
1973-75	43	24,300	32,863	50,539	135,851
1976-78	52	34,287	45,912	88,225	188,693
1979-81	56	43,800	60,135	159,976	259,242
1982-84	61	53,833	73,601	199,346	261,354
1985-87	64	72,301	96,647	245,407	290,285
1988-90	66	80,527	108,574	255,714	267,498
1991-93	53	82,955	110,642	266,537	248,595

Table 10. Continued.

Time Period	No. of Firms ^a	Processed Quantity		Processed Value	
		Product Weight	Headless Shell-on Weight	Current	Deflated ^b
		----- 1,000 lbs -----		----- \$1,000s -----	
----- Breaded -----					
1973-75	46	85,489	54,017	134,435	360,955
1976-78	38	79,032	50,154	170,874	365,938
1979-81	30	72,557	46,525	209,521	335,576
1982-84	26	78,273	49,040	291,051	380,221
1985-87	26	87,108	53,876	310,467	367,775
1988-90	26	104,051	66,277	300,205	315,076
1991-93	23	104,310	67,554	285,229	266,145
----- Specialty Products -----					
--					
1973-75	39	13,129	24,973	31,219	84,877
1976-78	34	12,830	26,510	41,232	88,586
1979-81	29	9,350	19,736	47,088	75,028
1982-84	25	6,965	14,020	39,944	52,081
1985-87	23	5,664	10,257	27,436	32,513
1988-90	21	5,901	8,277	27,073	28,907
1991-93	15	2,458		14,206	13,143

^a Note: summation of firms by product type for a given interval will not give the total number of firms in the industry (see Table 4) because many firms product more than one product.

^b The deflated value is given in 1990 dollars and is based on the Consumer Price Index.

As indicated, virtually all of the increase in Southeast shrimp processing activities during the 1973-93 period was based on increased peeling activities. Production of peeled shrimp, expressed on a product-weight basis, advanced from an average of 24 million pounds annually in 1973-75 to 83 million pounds annually in 1991-93 and the increase was consistent during the 21-year study period when examined in three-year intervals. With the exception of a sharp decline in number of firms during the most recent interval of analysis (from an average of 66 annually in 1988-90 to 53 in 1991-93), the increase in number of establishments peeling shrimp was also consistent during the 21-year period of analysis when examined in three-year intervals. The deflated value of peeling activities, after increasing from an average of \$136 annually in 1973-75 to \$290 million annually in 1985-87, declined to less than \$250 million annually by 1991-93.

Interval variation in the production of raw headless shrimp, to a large degree, reflected changes in reported Southeast shrimp landings (see Table 1). Such a finding is expected, given the fact that imports already arrive in a raw headless or more processed form. The deflated value of Southeast raw headless shrimp processing activities fell sharply after 1976-78, reflecting a declining deflated price of the processed product.

While the reported number of Southeast shrimp breeding establishments declined during the study period, the processed poundage increased to more than 100 million pounds (product weight) during the most recent two three-year intervals of analysis. While the processed breaded poundage increased substantially during the most recent intervals of analysis, the deflated value of the processed product fell sharply due to a decline in the processed per pound price.

The number of Southeast shrimp processing establishments producing specialty products fell sharply during the study period as did the processed poundage and deflated value. Much of the reduction was in canning activities which was likely in response to the substantial increase in canned imports.

SOUTHEAST U.S. 1991 SHRIMP PROCESSING ACTIVITIES

In 1991, a total of 130 shrimp processing firms were active in the Southeast U.S., according to NMFS records. About 95 of these firms reported processed shrimp sales in excess of \$200 thousand. To examine the Southeast shrimp processing sector at a greater level of detail than that permitted with the NMFS data, the authors selected a sample from these 95 firms to administer a detailed questionnaire. The questionnaire elicited information on monthly shrimp processing activities for the 1991 calendar year (see 1) for a copy of the questionnaire and additional discussion of the sampling techniques and survey). In total, 50 firms throughout the Southeast U.S. were included in the analysis. This represented more than 50% of the population of firms with 1991 reported processed shrimp sales in excess of \$200 thousand. Output among firms with processed shrimp sales less than \$200 thousand represented less than one percent on the industry total and were therefore not considered during the survey process.

For purposes of analysis, surveyed firms were divided into three categories: (1) those with processed shrimp sales of < \$5.0 million, (2) those with processed shrimp sales from \$5.0 million to \$15.0 million, and (3) those with processed shrimp sales > \$15.0 million. These categories are referred to in this paper as small, mid-sized, and large firms, respectively.

Finally, different weights were assigned to the three categories of firms based on the number of firms sampled in each category relative to the population of firms in that category. The data collected from the survey of processors was then extrapolated to the population, based on these weights, to provide an estimate of industry-wide processing activities in the Southeast Region. Hence, all results presented herein are industry estimates derived from the sample of firms (see 1 for details). Survey results are presented below.

Raw Material (i.e., Unfinished Shrimp) Supply

Raw Material Types. As noted, Gulf and South Atlantic shrimp processors utilize both domestic and imported shrimp to meet their processing needs. The types of shrimp that they use fall into four basic categories: (1) fresh/frozen heads-on shrimp, (2) fresh/frozen headless shrimp, (3) fresh/frozen raw peeled shrimp, and (4) other peeled shrimp products. The first category of shrimp, i.e., fresh/frozen heads-on, is essentially a domestic product. Fresh/frozen headless shrimp can be of either a domestic or imported origin. The two peeled categories are essentially imported products.

Estimated monthly purchases of domestic shrimp used in 1991 Southeast U.S. shrimp processing activities are reported in Table 11. In total, an estimated 143.1 million pounds of domestic heads-on shrimp and 56.9 million pounds of domestically produced headless shrimp were used in 1991 Southeast U.S. shrimp processing activities. This translates into a total of 148.5 million headless pounds when the heads-on shrimp are converted to a headless weight basis using a conversion factor of 0.63.

Table 11. Estimated Monthly Purchases of Domestic Shrimp Used in 1991 Southeast U.S. Shrimp Processing Activities.

Month	Heads-on	Headless	Total ^a
	----- 1,000 lbs -----		
January	3,172	2,421	4,420
February	1,847	1,681	2,844
March	2,501	1,701	3,277
April	3,520	2,013	4,230
May	24,725	4,918	20,494
June	29,419	5,397	23,931
July	17,378	8,034	18,983
August	11,097	9,107	16,099
September	11,174	6,438	13,478
October	16,657	6,798	17,291
November	14,166	4,948	13,872
December	7,455	3,445	9,584
Total	143,111	56,901	148,513

^a Heads-on poundage were converted to a headless basis using a conversion factor of 0.63.

Source: Compiled from (1).

Estimated monthly purchases of imported shrimp used in 1991 Southeast U.S. shrimp processing activities are reported in Table 12. As indicated, an estimated total of 103.7 million pounds of imported shrimp (expressed on a headless shell-on equivalent weight basis) was used in 1991 Southeast U.S. shrimp processing activities. This total was comprised of 59.5 million pounds of headless shell-on product and 34.5 million pounds of peeled product.

Table 12. Estimated Monthly Purchases of Imported Shrimp Used in 1991 Southeast U.S. Shrimp Processing Activities.

Month	Headless Shell-On	Peeled Raw and Other	Total ^a
	----- 1,000 lbs -----		
January	5,807	3,048	9,708
February	5,889	3,689	10,659
March	5,197	3,047	9,099
April	5,246	2,600	8,574
May	4,444	2,231	7,299
June	3,210	2,383	6,260
July	3,775	2,257	6,666
August	4,542	2,842	8,181
September	5,127	2,880	8,813
October	5,651	2,816	9,256
November	5,878	3,264	10,056
December	4,708	3,460	9,138
Total	59,474	34,517	103,709

^a Expressed on a headless shell-on equivalent weight using a conversion factor of 1.28 for peeled raw and other.

Source: Compiled from (1).

A comparison of the information contained in Tables 11 and 12 highlights three features. First, the information suggests that total industry purchases, expressed on a headless shell-on weight, equalled 244.1 million pounds in 1991. Forty-two percent of these purchases (103.7 million pounds) consisted of an imported product. Hence, while imported product constituted a minority of the raw material used in 1991 Southeast U.S. shrimp processing activities, import usage was sizeable.

A second feature highlighted by a comparison of the data in the two tables is that variation in domestic raw material purchases was substantially higher than that of the imported product. Total Southeast U.S. shrimp landings equalled 167 million pounds in 1991. About 85% of this total production (144.8 million pounds) was harvested from the Gulf

Region. All but 3.0 million pounds of the 148.5 million pound domestic purchases by processors (see Table 11) was estimated to be Gulf Region based, which indicates that essentially all of the Gulf Region production is used in processing activities, i.e., total Gulf Region landings equalled 144.8 million pounds and estimated processing activities from Gulf landed product equalled 145.5 million pounds. The variation in the monthly purchases of domestic shrimp essentially mirrors the seasonal nature of the Gulf Region shrimp fishery (see 1 for additional details).

A final feature highlighted by the data reflects the fact that variation in monthly purchases of imported raw material product was inversely related to the variation in purchases of the domestic raw material product. In particular, in the months when domestic raw material purchases were highest (May through August), import raw material purchases are lowest. This suggests that imported product was used seasonally when domestic product is more limited. As discussed by (1), this was particularly true among small (i.e., processed shrimp sales of < \$5.0 million) and mid-sized (i.e., processed shrimp sales of \$5.0 million to \$15.0 million) firms.

Annual shrimp purchases by size of firm for the 1991 year are presented in Table 13. As indicated, reliance on imports increased in relation to firm size. Among small firms, for example, 11% of total purchases consisted of an imported product (i.e., 3.8 million pounds compared to total purchases of 34.7 million pounds). The share increased to 20% among mid-sized firms and equalled 60% among large firms. This finding is expected, given the fact that the larger the operation, the more difficult it becomes to secure the needed raw material product from domestic sources on a year round basis.

Table 13. Estimated Raw Material Shrimp Purchases Used in 1991 Southeast U.S. Shrimp Processing Activities, by Size of Firm.

	Firm Size		
	< \$5.0 Mil.	\$5.0 - \$15.0 Mil.	> \$15.0 Mil.
Domestic Product:			
Heads-on (1,000 lbs)	38,342	63,192	41,567
\$/lb	1.28	1.27	1.68
Headless (1,000 lbs)	6,731	20,860	29,307
\$/lb	3.50	3.86	4.36
Total ^a	30,886	60,671	55,494
\$/lb	2.35	2.65	3.56
Imported Product:			
Headless shell-on (1,000 lbs)	3,841	12,445	43,236
\$/lb	3.23	3.33	3.44
Peeled (1,000 lbs)	0	1,968	32,550
\$/lb	---	3.90	2.87
Total ^b	3,841	14,964	84,900
\$/lb	3.23	3.28	2.85
TOTAL^c	34,727	75,635	140,394

^a Total domestic pounds and price per pound is presented on a headless basis.

^b Total imported pounds and price per pound is presented on a headless shell-on weight basis.

^c TOTAL equals combined domestic and imported raw materials expressed on a headless shell-on weight basis.

Source: Compiled from (1).

The information contained in Table 13 also suggests that the purchase price of the raw material increased in relation to firm size for comparable raw material products. For example, the domestic product (converted to a headless weight) was purchased by small firms at an average price of \$2.35 per pound compared to \$2.65 per pound among mid-sized firms and \$3.56 per pound among large firms. These differences, as will be discussed later, reflect two factors. First, the average size count of domestic shrimp purchases increased with firm size. Second, dependence on wholesalers increased with firm size which necessitates an additional marketing (cost) level.

Shrimp Sizes. For purposes of the study, processors were asked to identify the percentage of domestic and imported 1991 shrimp purchases that fell into the following headless size categories: < 30 count to the pound, 31-70 count to the pound, and > 70 count to the pound. Purchases of domestic and imported shrimp by size count was estimated based on the responses, the results of which are presented in Table 14.

Table 14. Estimated Purchases of Shrimp (Domestic and Imports) Used in 1991 Southeast U.S. Shrimp Processing Activities (given on a percentage basis) by Size of Firm.^a

	Firm Size			Total
	< \$5.0 Mil.	\$5.0 - \$15.0 Mil.	> \$15.0 Mil.	
----- Domestic -----				
< 30 ct. (%)	16.2	16.0	36.2	24.2
30-70 ct. (%)	37.6	41.6	36.1	38.7
> 70 ct. (%)	46.1	41.5	27.7	37.1
----- Imports -----				
< 30 ct. (%)	25.7	16.8	4.4	7.0
30-70 ct. (%)	53.9	58.6	59.7	59.3
> 70 ct. (%)	20.3	24.6	35.9	33.7

^a All count size data are provided on a headless weight basis.

Source: Compiled from (1).

As indicated, estimated industry use of domestic raw material supply was comprised as follows: < 30 count shrimp, 24%; 31-70 count shrimp, 39%; and > 70 count shrimp, 37%. As firm size increased, the use of larger domestic shrimp, as a percentage of the total by firm size, also increased. For example, < 30 count headless shrimp comprised only 16.2% of domestic shrimp usage among small firms compared to 36.2% among large firms. Shrimp > 70 headless count, on the other hand, comprised an estimated 46.1% of total domestic shrimp usage among small firms compared to 41.5% among mid-sized firms and only 27.7% among large firms.

Mid-sized shrimp (i.e., 31-70 count), as identified by the information contained in Table 14, dominated imported shrimp usage among all three categories of firms. Among small firms, 31-70 headless count shrimp represented 54% of their imported shrimp usage compared to almost 60% among mid-sized and large firms. However, use of small (> 70 headless count) imported shrimp among large firms accounted for more than 35% of their imported shrimp usage compared to less than 25% among mid-sized firms and small firms. As will be shown later, much of the imports of the > 70 count shrimp by the large firms is used in the production of breaded products and entails the peeled imports.

Procurement Source

Domestic procurement. Domestic shrimp supplies can be secured directly from the fishing fleet, from wholesalers/dealers, or from other sources. If secured directly from the fishing fleet, the product can be supplied from a processor's own boats/vessels or from other boats/vessels. Estimated procurement sources for domestic shrimp by Southeast processors in 1991 are outlined in Table 15. In total, an estimated 43% of the domestic supply (60.1

million pounds) was purchased directly from the fishing fleet while virtually all the remaining domestic supply was procured through wholesalers/dealers.

Table 15. Estimated 1991 Domestic Shrimp Procurement Sources
by the Southeast Shrimp Processing Sector.

Procurement Source	Firm Size			Total ^a
	< \$5.0 mil	\$5.0-15.0 mil	> \$15.0 mil	
	----- 1,000 lbs -----			
Supply From:				
Fishing Fleet	18,678	26,025	15,359	60,062
Own Boats (%)	6.4	5.3	4.4	5.4
Other Boats (%)	93.6	94.7	95.6	94.6
Wholesalers/Dealers	12,209	34,647	33,539	80,395
Other	0	0	259	259

^a The total given in this table (140.7 million pounds) is about 5% less than the 148.5 million pound total reported earlier due to non-response by some firms.

Source: Compiled from (1).

Direct dependence from the fishing fleet for domestic procurement declined in relation to firm size. Among small firms, for instance, 60% of domestic raw material supply was secured directly from the fishing fleet. For mid-sized firms, the share fell to 43%. The share equalled 31% among large firms. The observed decline in relation to firm size is expected, given the different types of operations. Many of the small and mid-sized firms are more seasonal in nature with processing activities heavily dependent on local fishing conditions. Large firms, on the other hand, are more dependent on a constant raw material supply throughout the year. Hence, they rely more heavily on wholesalers/dealers throughout the region to provide them with the needed domestic raw material.

The information in Table 15 also suggests that company fleets provide a relatively small share of the domestic supply secured directly from the fishing fleet. In total, only 5.4% of the domestic supply secured directly from the fishing fleet was estimated to come from company boats. The range was from 6.4% among small firms to 4.4% among large firms.

Import procurement. U.S. shrimp imports in 1991 equalled close to 540 million pounds (product weight) with more than 50 countries contributing to the total. Three countries, however - Ecuador, Thailand, and China - accounted for more than 50% of the total.

As noted, the estimated use of imported shrimp in Southeast U.S. processing activities equalled 103.7 million pounds (headless shell-on equivalent weight) in 1991. Estimated imports from China (32.4 million pounds) accounted for 31% of the total. Ecuadorian imports, estimated to equal 25.1 million, represented an additional 24% of the total. Imports

from other countries significantly contributing to 1991 Southeast U.S. shrimp processing activities included Thailand (9.6 million pounds, 9%), Honduras (6.4 million pounds, 6%), Indonesia (5.0 million pounds, 5%), Colombia (4.2 million pounds, 4%), and India (4.1 million pounds, 4%).

Processing Activities

Processing activities are reported in two sections. In the first section, information on processed quantities and the raw materials used in these processed quantities are examined. In the second section, processed prices are considered.

Processed Quantities. Monthly information was collected on the production of five processed shrimp products: (1) raw headless shrimp, (2) peeled raw shrimp, (3) peeled cooked shrimp, (4) breaded, and (5) "other" shrimp products. Based upon the responses by the fifty Southeast firms included in the analysis, monthly industry-wide production of these products was estimated, the results of which are presented in Table 16. As indicated, annual industry-wide production of raw headless shrimp equalled 77.0 million pounds; production of peeled raw shrimp equalled 78.5 million pounds (product weight); peeled cooked production equalled 12.9 million pounds (product weight); breaded production equalled 85.8 million pounds (product weight); "other" production equalled 4.2 million pounds (product weight). By and large, these numbers compare favorably with data maintained by the National Marine Fisheries Service. Unpublished NMFS data for 1991 indicated production of 94 million pounds of raw headless shrimp, 78 million pounds of peeled shrimp (raw and cooked), and 100 million pounds of breaded shrimp. Total production, expressed on a headless shell-on basis, equalled 265 million pounds. Hence, results from the current study suggest that raw headless and breaded production are underestimated while peeled production is overestimated (assuming the unpublished NMFS data are accurate). In total, the difference was about 6% (i.e., 249.0 million pounds compared to 265.0 million pounds).

Table 16. Estimated Monthly Production of Shrimp Products
by the Southeast Shrimp Processing Sector, 1991.

Month	Product Form ^a					Total ^b
	Raw Headless	Peeled Raw	Peeled Cooked	Breaded	Other	
----- 1,000 lbs -----						

January	3,744	2,674	1,136	6,924	66	12,766
February	2,903	2,094	943	8,236	69	11,865
March	3,323	2,136	787	8,902	113	12,667
April	3,628	2,663	841	6,515	98	12,109
May	6,825	11,772	1,006	6,309	888	28,151
June	7,238	14,185	1,245	6,072	810	31,193
July	10,329	8,254	1,026	6,056	625	26,731
August	11,283	6,071	1,166	7,239	190	24,778
September	8,302	6,000	988	7,529	204	21,712
October	8,767	8,993	1,051	7,910	410	26,521
November	6,655	8,470	1,290	7,199	463	23,837
December	3,975	5,139	1,401	6,912	256	16,704
Total	76,973	78,452	12,879	85,802	4,192	249,034 ^c

^a Poundage for individual products is given on a product-weight basis.

^b The total is given on a headless shell-on equivalent weight basis. The annual total of 249.0 million pounds is approximately 1.3% less than annual purchases of raw input, expressed on a headless shell-on weight, of 252.2 million pounds. Much of the difference likely reflects the average conversion factors used in deriving the shell-on weight from the different product weights.

^c Horizontal summation of annual totals by product form will not yield the annual total which is given on shell-on equivalent weight.

Source: (1).

The information in Table 16 suggests considerable variation in raw headless, peeled raw, and "other" production. Variation in the production of peeled cooked and breaded output was much less evident. The reason for the monthly variation in the production of these three products and lack thereof in the production of the other two products can be ascertained from the information contained in Table 17.

Table 17. Information on Southeast Industry-Wide Production of Different Processed Products.

	Product Form				
	Raw Headless	Peeled Raw	Peeled Cooked	Breaded	Other
Total Output (1,000 lbs)	76,973	78,452	12,879	85,802	4,192
Raw Material (1,000 lbs)					
Heads-on	12,087	118,160	852	0	12,906
Heads-off	70,159	15,209	8,179	20,285	3,268
Peeled	-----	-----	6,050	28,139	437
Supply Sources (%)					
Domestic	78.1	84.8	3.9	0.8	78.6
Imports	21.9	15.2	96.1	99.2	21.4
Size Count (%)					
< 30 ct	40.9	3.8	9.9	10.7	0.0
30-70 ct	50.2	37.3	79.6	49.1	50.2
> 70 ct	8.9	58.9	10.5	40.2	49.8

Source: (1).

As indicated, the output of raw headless shrimp, peeled raw shrimp, and "other" shrimp was highly dependent on domestic shrimp landings which are also highly seasonal (see Table 11 for monthly domestic purchases). About 85% of industry-wide production of peeled raw shrimp in 1991 was found to be derived from domestic raw product while close to 80% of the raw headless and "other" shrimp output was domestic raw material product based. In contrast to this finding, only 4% of the peeled cooked product and less than one percent of the breaded product was derived from domestic raw material.

The information in Table 17 also indicates that the overwhelming majority of domestic large shrimp production (i.e., < 30 count headless) was used in raw headless processing activities. While size of shrimp used in the production of the different product forms was not differentiated between domestic and imported shrimp, the aforementioned statement can be substantiated with a few facts. First, all Gulf of Mexico production appears to be processed. Second, domestic landings are basically used in the production of three product forms: raw headless, peeled raw, and "other" shrimp products. However, very little large shrimp is used in the production of the latter two products. Hence, essentially all domestic production of large shrimp must go into raw headless processing activities. Using the same logic, it can be surmised that the overwhelming majority of domestic small shrimp (i.e., > 70 count) is used in peeled raw processing activities and to a much lesser extent the production of "other" shrimp products (due to relatively small total production of "other" processed shrimp

products). Domestic landings of mid-sized count shrimp appear to be distributed somewhat evenly between raw headless and peeled raw activities.

Peeled raw material, as indicated, was used primarily in breaching activities and, to a lesser extent, peeled cooked activities. The heads-on raw material was overwhelmingly used in the production of peeled raw output.

Estimated raw headless production of processed shrimp by firm size is presented in Table 18. Large firms accounted for 50% of annual production compared to 33% among mid-sized firms and 17% among small firms. Domestic shrimp accounted for 76% to 80% of raw headless output among all three categories of firms. Large firms, however, used a much higher percentage of < 30 count shrimp in their raw headless processing activities than do either the mid-sized or small firms. This finding relates directly to the relatively large purchases of < 30 count domestic shrimp by the large firms (see Table 14).

Table 18. Information on Southeast Industry-Wide Production of Raw Headless Processed Shrimp by Firm Size, 1991.

	Firm Size			Total
	< \$5.0 mil	\$5.0-15.0 mil	> \$15.0 mil	
Total Output (1,000 lbs)	13,207	25,107	38,659	76,973
Raw Material (1,000 lbs) ^a				
Heads-on	5,081	651	6,357	12,089
Headless	10,224	25,265	34,670	70,159
Supply Sources (%)				
Domestic	75.9	76.9	79.6	78.1
Imports	24.1	23.1	20.4	21.9
Size Count (%)				
< 30 ct	22.6	37.4	49.4	40.9
30-70 ct	60.6	54.7	43.7	50.2
> 70 ct	16.8	7.9	6.9	8.9

^a Summation of heads-on poundage (x 0.63) and headless poundage does not necessarily equal total output. Some raw material may be lost in processing.

Source: (1).

Estimated peeled raw shrimp processing activities by size of firm is presented in Table 19. While domestic raw material served as the basis for the majority of raw peeling activities among all firms categories, there existed a generally lessening of the dependence in relation to firm size. Among small firms, for example, domestic shrimp constituted an estimated 97% of the raw input used in the production of peeled raw shrimp. Among large firms, the share was less than 80%. In addition, large firms used a much lower percentage of small shrimp

in their peeled raw activities than did either the small or mid-sized firms. To some extent, this may reflect the higher import usage in the production of peeled raw product in relation to firm size.

Table 19. Information on Southeast Industry-Wide Production of Peeled Raw Shrimp by Firm Size, 1991.

	Firm Size			Total
	< \$5.0 mil	\$5.0-15.0 mil	> \$15.0 mil	
Total Output (1,000 lbs)	17,096	32,895	28,460	78,451
Raw Material (1,000 lbs)				
Heads-on	30,877	51,830	36,153	118,160
Headless	355	5,411	9,442	15,209
Supply Sources (%)				
Domestic	97.9	83.9	77.8	84.8
Imports	2.1	16.1	22.2	15.2
Size Count (%)				
< 30 ct	2.6	2.4	6.2	3.8
30-70 ct	38.9	29.2	45.7	37.3
> 70 ct	58.5	68.4	48.1	58.9

Source: (1).

Breading activities by size of firm are presented in Table 20. As indicated, the vast majority of breading activities was conducted by large firms. Both groups of firms that processed breaded shrimp used essentially all imported raw material in their breading activities. Mid-sized firms used primarily headless shell-on shrimp in their production of breaded shrimp while large firms depended more on peeled raw material. Mid-sized firms also used a larger size count shrimp, on average, in their production of breaded products. Less than 20% of the raw product used by mid-sized firms was < 70 count shrimp compared to more than 40% among the larger firms. This likely relates to the fact that peeled imported shrimp, of which large processors are more dependent upon heading operations, tends to be a smaller size count than headless shell-on imported shrimp.

Table 20. Information on Southeast Industry-Wide Production of Breaded Shrimp by Firm Size, 1991.

	Firm Size ^a		Total
	\$5.0-15.0 mil	> 15.0 mil	
Total Output (1,000 lbs)	8,112	77,690	85,802
Raw Material (1,000 lbs)			
Headless	2,840	17,445	20,285
Peeled	1,079	27,060	28,139
Supply Sources (%)			
Domestic	0	0.8	0.8
Imports	100	99.2	99.2
Size Count (%)			
< 30 ct.	18.7	9.9	10.7
30-70 ct.	61.7	47.8	49.1
> 70 ct.	19.6	42.3	40.2

^a No firms < \$5.0 million were surveyed that produced breaded shrimp products.
Source: (1).

The small number of firms producing peeled cooked and other shrimp products limits meaningful discussion of their products with respect to firm size. Peeled cooked products were overwhelmingly produced by large firms (an estimated 94% of the total). While "other" products were produced by all firms categories. The number of firms in each category was extremely small (one or two).

Prices of Processed Products. Prices received by processors for the different product forms are reported in Table 21. The weighted average industry price reported for raw headless processed shrimp was \$4.56. Small firms received a much lower price per pound (\$3.74) than did either the mid-sized firms (\$4.78) or large firms (\$4.56). The relatively low price received by the small firms clearly reflects the smaller average size shrimp used in their production of a raw headless processed product (see Table 19). More difficult to explain is the lower price received by large firms when compared to mid-sized firms. One explanation is that some of the large firms that produced only raw headless product acted only as custom packers. The custom packers worked only on commission. As such, the reported sales price is "artificially" low, since profits are not included.

Table 21. Estimated Sales Prices Related to Different Processed Shrimp Products, 1991.

Product Form	Firm Size			Weighted Average
	< \$5.0 mil	\$5.0-\$15.0 mil	> 15.0 mil	
	----- \$/lb -----			
Raw Headless	3.74	4.78	4.56	4.56
Peeled Raw	3.12	2.87	3.75	3.25
Peeled Cooked	--- ^a	---	---	5.44
Breaded	---	3.47	2.90	2.95
Other	---	---	---	6.52

^a Not estimated or not applicable.

Source: (1).

The industry-wide peeled raw processed price was found to equal \$3.25 per product weight pound. The relatively low price received by mid-sized firms (\$2.87) reflects their high usage of > 70 count shrimp in their processing activities (see Table 20). Similarly, the relatively high price received by large firms (\$3.75) reflects their higher use of larger shrimp.

Peeled cooked shrimp received an average price of \$5.44 per product weight pound compared to \$2.95 for the breaded product and \$6.52 for "other" processed products. Mid-sized firms received a higher price than did large firms for the breaded product reflecting, in part, larger shrimp used in production.

END NOTES

- ¹ The 124 reported annually in 1991-93 was significantly higher than that reported in the most recent survey year (117).
- ² The product weight includes the meat weight of any shrimp used in the processing activities, plus any additional ingredients that may be added such as breading materials, plus shell weight if appropriate (such as in the case of frozen raw headless).
- ³ The National Marine Fisheries Service database use in the current processing study includes information on the average number of workers per firm involved in processing activities. Many shrimp processing firms, however, also produce other species and processing employment by species is not available on a per firm basis. To analyze shrimp processing activities per worker, therefore, the authors included in the analysis only that group of firms primarily engaged only in shrimp processing activities (defined as > 95% of the value of processed sales were shrimp based). In general, this method of analysis included from 60% to 70% of the total number of Southeast shrimp processing establishments on an annual basis.

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DOES SEAFOOD SCIENCE HAVE A ROLE IN FISHERIES MANAGEMENT: A CASE STUDY OF THE PACIFIC WHITING FISHERY

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INTRODUCTION

Optimum utilization of fishery resources is a critical issue for the industry and seafood researchers. There is a general consensus that wild-caught marine fishery harvests have peaked and the majority of the commercially important fisheries are at their maximum sustainable yields (de Wilde and Kamstra 1995). Maximum utilization and increased economic benefits to the industry can still occur through reduction of postharvest fishery losses, increased recovery of food grade raw material from fish, and harvesting when fish are in their optimal physiological state in terms of seafood quality and potential processing yield. Most commercial marine species undergo physiological changes that affect the potential quality and processing yield. The European sardine undergo tremendous seasonal changes in lipid content varying from 1.6% to 22.4% lipid content on a whole body basis (Ackman 1995). Salmon rapidly deteriorate during spawning migration which greatly affects flesh quality and potential markets. Several species undergo specific spawning periods after which the muscle becomes noticeable soft and has a high moisture content. Love (1988) has described several physiological changes that occur during spawning and their effects on the "eating quality" of fish. There has been little effort to incorporate these changes in physiological conditions into fisheries management schemes. This paper will discuss the need for using intrinsic fish quality characteristics as a management tool. The Pacific whiting fishery is used as an example of how fisheries management plans can incorporate seafood science data to increase economic benefits and help conserve the resource.

Pacific whiting (*Merluccius productus*) is the largest fishery resource off the Pacific Northwest Coast (excluding Alaska). Since 1990 through 1996 the harvests have averaged approximately 207,000 metric tons (mt) with lows of 142,000 mt (1993) and highs of 260,000 mt (1994) (Radtke, 1995). The fishery has evolved from one dominated by foreign

and joint venture (JV) operations in the 1980s, to a split at-sea and shoreside operations during the 1990s. In 1996, 40% of the fishery was taken by shoreside interests while 60% was harvested by an at-sea component which consists of large factory trawlers and mother-ships that act as processors. Shoreside processing of Pacific whiting has grown from 8,115 mt in 1990 to a present harvest of 85,000 mt. This ten-fold increase is the result of utilization of Pacific whiting for surimi production. There are four surimi plants in Oregon that use from 200,000 to 500,000 lbs of Pacific whiting per day as raw material. Although several products, such as frozen H&G, IQF fillets and minced blocks, are made from whiting, more than 80% goes into surimi production. The main stock of Pacific whiting spawns off the Baja California coast in the winter and slowly migrates up the California coast in the Spring (Wilkins 1992). By March, the fish can be found in large schools in near-shore waters from Northern California to south of Vancouver Island, Canada. The larger factory trawlers that enter the fishery will harvest their quota within a two-three week period. The onshore fishery, depending on the size of the quota and the number of vessels fishing, will last 60 to 100 days.

The Pacific Fisheries Management Council (PFMC) develops the management plan for the Pacific whiting resource in the U.S. This involves a variety of regulatory options including setting the opening dates of the season as well as allocating the annual allowable domestic harvest among competing fishing and processing sectors. The opening dates for the U.S. fishery have traditionally been April 15 which accommodates the needs of the offshore fleet to operate between the Alaska A and B seasons. The allocation decision (between onshore and offshore interests) usually involves a choice among alternatives based on economic and social benefits (e.g. employment) that each sector would generate. These decisions are made to satisfy three broadly defined goals of the Pacific Coast Groundfish Fishery Management Plan. These are: 1) conservation of the resource, 2) maximum economic value, and 3) efficient utilization. In conducting their analysis, policy makers have largely ignored the intraseason variation in the characteristics of the raw product. This variation can have a significant impact on product quality and ultimately, socioeconomic benefits of the fishery.

There is much anecdotal and research information concerning Pacific whiting quality. It is primarily known as a soft-flesh whitefish that can have severe texture problems if not handled properly (Peters et al. 1995). There have been anecdotal reports that during the JV operations, European buyers would not accept fish harvested before June due to quality problems. The quality issue is centered on two concerns, one biochemical and the other physiological. Pacific whiting is known for having high levels of protease enzymes in their muscle tissue. These proteases are associated with myxosporidean parasites that infect the flesh. Although the parasites are not a human health problem their presence and that of the protease can effect the final product quality. Onshore harvests of Pacific whiting, prior to 1990, were carried out by bottom trawlers that primarily captured other groundfish species (i.e. rock fish) and often fished over a 3-4 day period. Due to texture problems, efforts were made to capture whiting on the last days of the trip and on-board handling of whiting was less than optimal.

With the advent of the surimi industry, Pacific whiting became a target fishery and the industry began to make changes. Most boats altered their hulls and had refrigerated sea water (RSW) systems or champaign ice (seawater, ice and bubbled air) systems installed. Research at the OSU Seafood lab showed that time and temperature were critical factors for production of high quality surimi (Morrissey et al. 1992). Researchers recommended that the fish be brought down to $<4^{\circ}\text{C}$ and be processed within a 24 hr period. Trips were often accomplished within 12 hrs and fish were stored either in the vessels in refrigerated systems or on shore based tanks that were refrigerated. Research with different protease inhibitors showed the correct combinations that could be used for inhibiting the proteases that remained in the flesh (Morrissey et al. 1993). Additional research identified the protease enzymes responsible for tissue softening (Seymour et al. 1994; An et al. 1994), loss of protein in surimi processing (Lin and Park, 1996a) and the effects of in-line washing on surimi yield and quality (Morrissey et al. 1995; Lin and Park 1996b). The OSU Seafood Laboratory worked closely with the industry in determining the factors in the Pacific whiting fishery that affect final surimi quality. Work with new "expert systems" such as neural networks and M-5 induction gave insights to researchers and the industry about biological, harvesting and processing factors that effect final product quality (Peters et al. 1996).

On-board handling and processing factors can be changed to optimize quality if the fishermen and processors agree that the costs of the changes are economically worthwhile. The intrinsic biological factors, as they relate to seasonality, can only be changed through fishery management decisions that reflect harvests dates, allocations, etc. The purpose of this study was to determine the changes in intrinsic quality parameters that take place during the Pacific whiting season and determine how they effect critical factors such as yield and product quality. If these factors are important for the economic health of the industry, then suggestions can be made to the fishery management council that would reflect a maximization of profits to the industry without compromising the biological health of the fishery.

METHODOLOGY

Development of Pacific Whiting Model

The fishery is modeled using non-linear mathematical programming. The type of model involves choosing the time of harvest which maximizes social benefits subject to biological dynamics and economic conditions (Clark 1990; Onal et al. 1991). One of the main objectives of the model is to maximize the Net Present Value (NPV) to the industry by incorporating biological, economic and intraseason product quality variation. Relationships between product characteristics and prices, costs, recovery rates, and production practices are determined using intraseasonal data. For the Pacific whiting fishery, the model is based on the final product form - surimi, which is a graded product and one of only a few fisheries-based products which have an established structure for quality determination. The quality characteristics of surimi determine its price and the production formulas for several hundred surimi-based final products. The other product forms (fillets, H&G, etc.) are not marketed

using a consistent set of quality parameters nor have they experienced significant inseason price variation.

Surimi quality is typically identified by quantifiable levels of certain quality parameters. Together these characteristics are used to define its grade, and ultimately the price of the product. These include gel strength, whiteness, and moisture content. Seasonality is typically incorporated into a model using a dummy variable, however, in our analysis, seasonality is implicitly incorporated through changes in the properties of surimi which result from variation in intrinsic quality (i.e., flesh composition). The equations specifying this relationship were estimated as an SUR modeling system (i.e., the surimi characteristics were regressed against raw product characteristics in order to relate intrinsic quality to surimi quality)(Larkin and Sylvia 1996a). Only gel strength was found to be significant at the 5% level. Using the coefficient, a reasonable 25% seasonal increase in gel strength would produce a \$0.10 per pound, or 16%, price increase. The data utilized was obtained during a period of relatively stable supply and demand conditions. Obviously supply and demand cannot be expected to remain constant in the long-run, however, our goal for this analysis is only to incorporate price variation to the extent that it is affected by the collective changes in raw quality (Larkin and Sylvia 1996b).

Data Collection

Several data sources were used for this model. Weight and length data by age were obtained from 1986-1988 (PFMC 1992). The Oregon State University Seafood Laboratory (OSU-SL) provided information on weights, lengths, and proximal content for 1992-1994 seasons. Pacific whiting fish samples were taken each week over a three year period. For each analysis 20 fish were chosen at random and transported on ice to the OSU-SL. Proximate composition (protein, moisture, lipid and ash) were done on Pacific whiting fillets by standard AOAC methods (AOAC 1990). Data sources for earlier years (mid 1960s) and similar species (cod, pollock) confirm the stability and absolute values of our data (Nelson et al. 1985; Alaska Fisheries Development Foundation 1991). Annual average product recovery rates were obtained from NMFS and inseason rates were provided by two private firms on a confidential basis. Data for the determining the relationship of price to surimi characteristics were received from four distinct, but confidential, sources which enabled model validation. Remaining price and cost data was obtained from NMFS and correspond to the estimates used by the resource managers (PFMC 1993).

Biological Information

The biological component of the data uses an estimate of the initial population, and incorporates annual recruitment, migration, natural mortality, and fishing selectivities to determine the annual population, spawning biomass, fishing mortality, and sustainable harvest levels. The annual allowable U.S. harvest is allocated into monthly catch. The data for the biological model was obtained from the National Marine Fisheries Service (NMFS) which develops the stock assessments for management of the Pacific whiting fishery (Dorn et al. 1993).

Economics

In our model, each sector realizes different product recovery rates, costs, and in some cases, prices. In addition, each sector specializes in the production of different products; the offshore sector concentrating in surimi production while the onshore sector produces surimi and some fillets and H&G products. Each sector differs in its maximum daily capacity and utilization of waste. The shoreside industry has spawned several by-product industries such as the production of fishmeal, protein hydrolysates, and fertilizers. Product recovery rates vary by product form (surimi, H&G, fillets and fishmeal) and are multiplied by the gross weight of the fish used to arrive at final production quantities. These rates are also used as indicators of the efficiency of the industry or comparison of the efficiency of competing harvest sectors.

Seafood Science

The general form of the intraseason product recovery rates (a.k.a. production, yl_d),

$$yld_{m,s,t} = f(X_1, X_2) = f(cf, wl, pro, moi, fat)$$

assumes that yields are determined by both fish size (X_1) and flesh composition (X_2), where fish size is described by either the condition factor (cf) or the weight-length ratio (wl) and flesh composition consists of the percentage of weight accounted for by protein, moisture, or fat (pro , moi , and fat , respectively). In particular, a larger fish (e.g., heavier or "plumper") can either increase the recovery rate (processing equipment is generally able to extract more from larger size fish) or decrease it (if size is a result of increased gonadal tissue). Similarly, improvements in the composition of the flesh can either increase recovery rates (as protein content and quality are positively related) or decrease rates (as moisture content and quality are inversely related). Partial correlation analysis identified which variables describe the most variation in each of the production yields. This system of linear equations was estimated using SUR (Larkin and Sylvia 1996a).

RESULTS

Protein content in Pacific whiting fillets increased and moisture content decreased as the season progressed (Fig. 1). The normal whiting season begins April 15 and ends once the quota is captured. At the beginning of the season the moisture content is usually close to 84% as the fish are recovering from their post-spawning migration. This gradually decreases to 81% by the end of July. The protein content in the fillets increased as the season progressed from a low of 15% to 18% by August. This indicates improved condition of the fish and better overall quality as well. The surimi process requires washing the minced flesh to concentrate myofibrillar protein. Optimum surimi production would take place during the period when there is maximum protein in the flesh. The weight-length ratios as tracked over time. This data also shows that the condition of the fish improves as the season progresses and the fish put on weight.

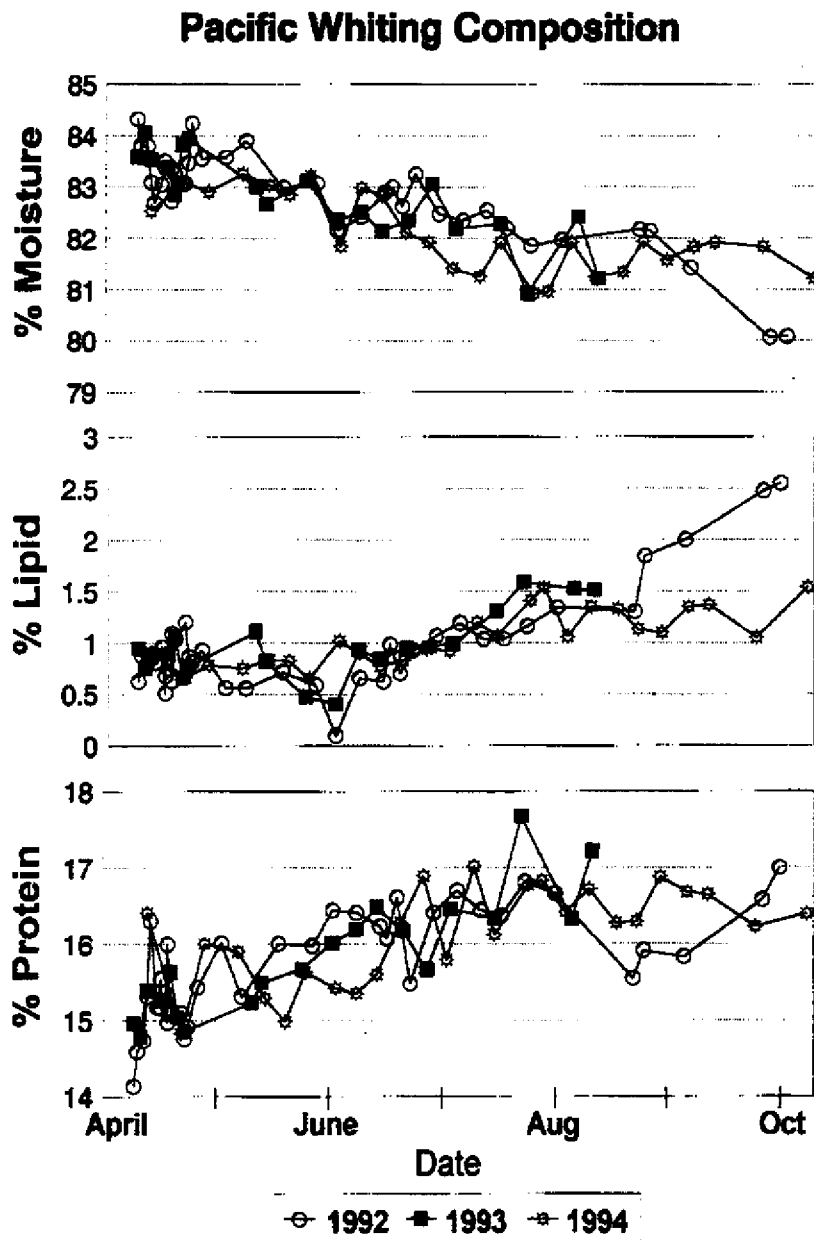


Figure 1. Moisture, lipid and protein content of Pacific whiting over the 1992-1994 harvest seasons

The optimized model described in the methodology section maximizes the value of the fishery over time by determining when, where, by whom, and how much is harvested. The optimal management plan (i.e., the "proposed" plan) is compared with the current management practice which ignores seasonality (i.e., the "standard" plan). More importantly, the model describes the differences between plans and quantifies the individual effects of

changes in weight, product recovery rates (via intrinsic quality changes, i.e., changes in size and flesh composition), and market prices for the final goods.

Results show that the inseason timing of harvests are significantly affected by having the model include seasonality. Incorporating inseason changes shows that delaying the harvest to coincide when the fish are at optimum quality increases Net Present Value (NPV) 117%. These results are summarized in Fig. 2. Under standard management, the offshore sector extracts its quota in April (usually in 2-3 weeks) and, on average, the season closes in July for the onshore component. Under the new proposed management plan the onshore season would not open until July and the offshore sector would harvest in October.

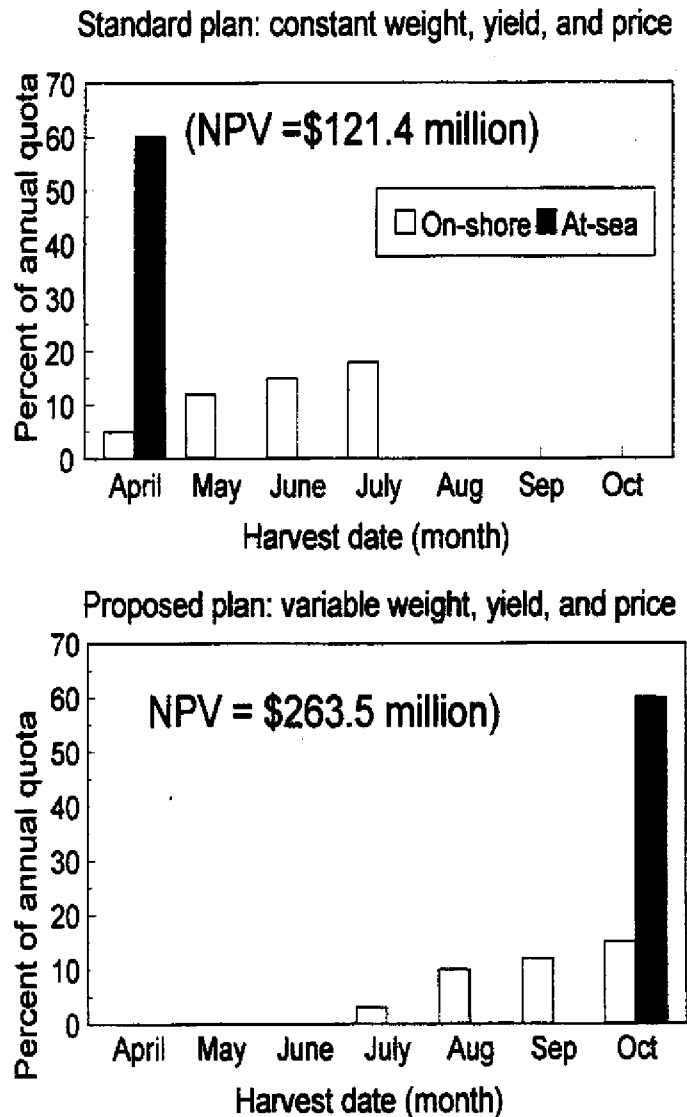


Figure 2. Harvest schedules for Pacific whiting as described in the existing standard plan and the proposed plan. (NPV = Net Present Value.)

The individual monetary effects that would result from the new fishing schedules are shown in Fig. 3. This figure describes the increase in NPV, that is, the difference between the standard and the proposed plan. The recovery rate (yield) effect dominates by accounting for 38% of the increased NPV. Inseason price and weight changes contribute 25% and 6%, respectively. These individual effects were determined by systematically allowing each component to vary while holding the remainder constant and recording the change in NPV (Sylvia et al. 1996).

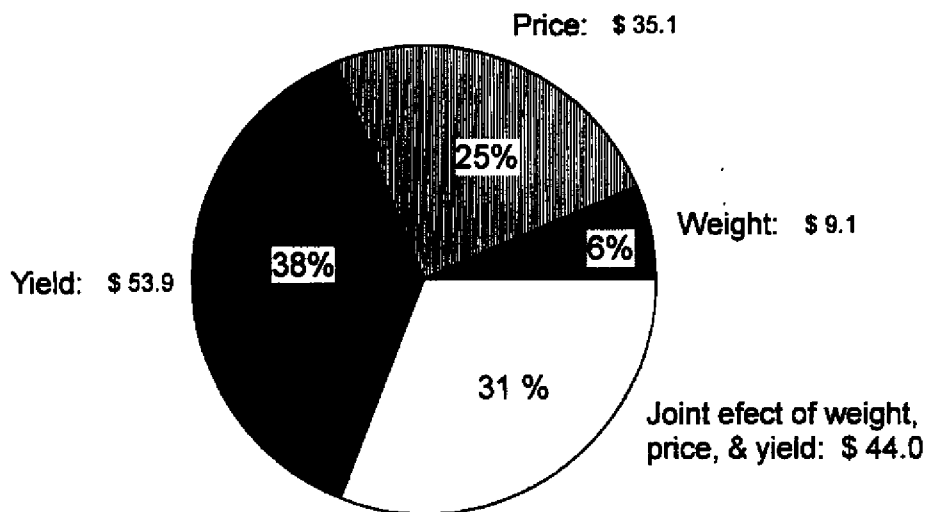


Figure 3. Relative percent contribution of weight, price, and production yields to increased NPV (\$ million) resulting from introduction of seasonal variability.

It is possible to separate yield and price effects even further by examining the changes in the condition of the fish at the time of capture, i.e. fish size and flesh composition. For example, as the moisture content of the fish declines throughout the season (from approximately 84% to 81%), both production yields (of surimi and fillets) and surimi characteristics (including water content, whiteness, and gel strength) are affected. This is also true for protein and fat content. Seasonal changes in the intrinsic characteristics provide the critical link, in terms of effective management, between biological and economic components.

IMPLICATIONS ON FISHERIES MANAGEMENT

Clearly, the economic goals of the Pacific whiting fishery plan are best met if the season opening is delayed until June-July. It is important to consider the implications of this delayed opening on other factors such as conservation of the resource. The standard management plan is compared with the proposed plan on the basis of the stated goals of the PFMC Groundfish Management plan: 1) conservation of the resource, 2) economic value, and 3) efficient utilization. Results are summarized in Table 1.

Table 1. Comparison of management goals under the standard and proposed plans.^a

U.S. management goals	Management plans		
	Standard	Proposed	Relative change
(1) Conservation			
1a) number of fish harvested	513.7 million	461.7 million	- 10%
1b) weight of fish harvested	225,100 t	253,000 t	+ 12%
1c) size of spawning biomass (year 10)	1015 million	1028 million	+ 1%
1d) relative size of harvest (harvest/spawning biomass)	46.6%	41.5%	- 11%
(2) Economic value			
2a) net present value (NPV)	US \$121.4	US\$263.5	+ 117%
2b) property rights allocation			
onshore	46%	37%	- 19%
offshore	54%	63%	+ 16%
(3) Utilization			
3a) product recovery rates (yld)			
surimi	14%	17.4%	+ 24%
headed and gutted	56.4%	61.4%	+ 9%
fillets	23.5%	27.2%	+ 16%
meal and oil	9.8%	11.0%	+ 12%
3b) output quantities			
surimi	28,400 t	39,100 t	+ 37%
headed and gutted	8,000 t	8,000 t	0%
fillets	5,000 t	5,000 t	0%
meal and oil	27,900 t	24,100 t	- 16%

^a Values represent the average of annual statistics unless otherwise noted. If there is a seasonal component, the annual measure is the seasonal average based on when the harvest occurred.

No single measure can adequately represent the conservation of a resource. We use four separate measures to understand the possible implications to the ecosystem of the proposed management plan. The new proposed plan offers greater conservation since both the absolute and relative number of fish harvested declines (1a and 1d, respectively). In addition, the total annual harvest quota increases, that is, fewer but heavier fish are harvested. Perhaps more importantly, the size of the spawning biomass remaining at the end of the planning period is not compromised (1c). For the second goal, NPV increases under the proposed plan (NPV is our interpretation of the economic management goal). Alternatively, one could compare changes in employment or other measures of economic value to society (e.g., the relative

change in property rights, 2b). In terms of utilization of the resource, average recovery rates increase under the proposed plan (3a). Output of surimi increases as property rights are shifted to the offshore sector (which specializes in surimi production), while production of meal declines. In addition, the onshore production of surimi also increases under the proposed plan (resulting from the seasonal price effect). The quantity of fillet and headed and gutted product does not change since both current production capabilities and market opportunities are limited.

SUMMARY

Seafood science can play an important role in management of a fisheries. There has been ample documentation of compositional changes in many commercially harvested species. These changes in moisture, protein, lipid content will affect final product quality, yields, and economic returns to the industry. This paper showed how variations on intraseason intrinsic quality can be an important key for successful development and management of many wild-stock fisheries. An interdisciplinary model for the Pacific whiting fishery demonstrated how seasonal changes in the raw product quality (i.e., fish weight at harvest and relative size and composition of the flesh) influence the economics of the fishery and its management. In particular, variations in weight directly affect the harvest quantities and variations in the relative size and proximate content impact production yields, product quality and product price.

For many species, management has disregarded the inseason timing of harvest in order to focus on the issues including allocating the annual quota among the competing harvest sectors. Failure to consider inseason intrinsic variability, however, results in sub optimal management of fast-growing or rapidly changing stocks. The result is decreased benefits to society and potentially the ecosystem. More importantly, management goals may not be mutually exclusive if harvest policies are dictated by the characteristics of the individual fish; that is, goals such as conservation, efficiency, and utilization may often be complementary.

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THE IMPACTS OF FEDERAL FOOD SAFETY INITIATIVES ON THE AQUACULTURE PRODUCER-PROCESSOR HACCP JUNCTURE

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Food safety and related foodborne illnesses are important issues to consumers. Food producing sectors have recognized the need to link their products with safety-quality standards and controls to gain better consumer confidence in the marketplace. Two critical issues that will have a significant impact on the future successes of food animal production in the U.S. are food safety and price competitiveness, in addition to new market and product alternatives. A 1996 survey of U.S. consumers found that 80% of respondents were concerned about food safety (1). The public health emphasis is to avoid disease based upon a lifetime of chronic exposure to residues. International efforts to harmonize safety standards and guidelines for aquatic food products are causing changes that have farm (production) through processing, distribution and retailing to fork (consumer) implications.

Industry, as evidenced by quality assurance programs (QAPs) and Hazard Analysis and Critical Control Point (HACCP) initiatives, realizes that one critical key to growth in existing and new markets is to develop a quality and safety ethic from farm to market. There are potential human health risks when contamination occurs in any food product. The main health issue usually associated with aquacultural products is the positive dietary and health benefits (2). Of the public health risks associated with aquaculture, the most serious are foodborne illnesses associated with the consumption of raw molluscan shellfish (3). Even though domestic finfish are low safety risk products, there is a continuous need for the industry to institute adequate safety control measures to prevent potential public health risks and maintain high consumer acceptance for all farm-raised products.

Before 1989 the U.S. aquaculture sector received minimal regulatory attention from Federal agencies with public health missions. After rapid growth during the 1980's, this sector however gained increasingly important recognition as a source of consumer aquatic food products. The aquaculture industry had been using animal drugs and chemicals in a manner that was not acceptable and below the requirements and standards of other animal

protein producing industries. If the aquaculture industry was serious about developing into a significant production system, changes were needed. Since 1990, FDA inspection, surveillance and compliance programs regarding the sale and potential use of unapproved drugs and chemicals have become more active and similar to those for other animal agriculture food sectors (4). Concurrently, educational initiatives, through producer QAPs which address potential human food safety issues related to HACCP principles (5,6,7), have been developed that promote the proper use of therapeutants and chemicals. There are only five drugs approved for the diverse mix of aquaculture species in the market, yet there is a need for other drugs to maintain the health of commercial species. Increasing resistance of fish pathogen strains to approved antibiotics is also aggravating health management options with therapeutants.

With the seafood HACCP regulations of the U.S. Food and Drug Administration (FDA) which mandated seafood HACCP-based inspection regulations (8), the aquaculture processing sector and importers of farm-raised aquatic food products are developing HACCP plans to comply with the new requirements which become effective December 18, 1997. HACCP provides the aquaculture industry with an important food safety control tool to prevent problems from ever reaching consumers - the ultimate beneficiary (9). Seafood processor HACCP plans may require producers to provide evidence and assurances for pre-harvest product safety.

The integration of voluntary producer-based QAPs and mandatory processor HACCP plans can facilitate the adoption of safety control measures for the first potential critical control point (CCP) - receiving raw, farm-raised products. There is a need for practical, reasonable monitoring options based on historical data and available testing tools, adequate recordkeeping systems and adherence to QAPs with verification protocols that producers, processors and regulators understand and accept. More communication is needed between the three parties to establish acceptable options, procedures and expectations that address compliance issues.

Processors will need to be assured that they are receiving farm-raised products that are safe and wholesome through knowledge of the production history of the products they purchase. The need for more information may change their contractual requirements with their suppliers. The seafood HACCP regulations (8) and the Fish and Fisheries Products Hazards and Controls Guide (the Guide) (10) address the safety issues of farm-raised products. However, some of the compliance requirements are unclear and are prone to different interpretations by industry and inspectors. Aquaculture drug use issues linked to producer QAPs and the implementation of HACCP by processors have proceeded on separate, but parallel tracks. Voluntary on-farm (pre-harvest) and mandatory processor HACCP-linked food safety programs are actually interrelated. In December 1997 these two tracks will converge when processors actually execute HACCP plans that are scrutinized by government inspectors for completeness and correctness.

This paper provides examples of the impacts of Federal initiatives and resultant public and private sector actions that can effectively bridge the HACCP food safety requirements to the processors who process a wide variety of food species. An historical overview is also presented including information on baseline residue sampling data at the farm level. Recommendations are made that serve as guidance to producers, processors and regulators to facilitate an effective transition into this new HACCP regulatory system.

METHOD AND MATERIALS

Numerous documents were reviewed to identify issues associated with producers, processors and regulators regarding the mandatory seafood HACCP inspection regulations. These included FDA's seafood HACCP regulations (8), the Guide (10), various FDA reports, presentations and field compliance programs, and existing producer QAPs. Additionally, data on sampling and testing of aquaculture sites and products for drug, chemical and pesticide residues were evaluated.

The importance of topics was identified through discussions and interviews with both industry and regulatory representatives. These perspectives assisted in evaluating the need for the development of options and recommendations that can assist all public and private sector parties involved with the new mandatory seafood regulations. With strategic approaches that address unresolved or uncertain issues now, much confusion and undue burdens can presumably be prevented when the seafood HACCP regulations are enforced. Collaboration and understanding will be required by the processors developing HACCP plans, the producers who may be impacted because of new purchasing requirements linked to potential pre-harvest safety concerns, and the regulators who will be inspecting firms and enforcing the regulations.

The recommendations presented were also reviewed by both public and private sector representatives to gain a clearer understanding of the critical issues and options for their resolve. Our intent is to increase the awareness of critical issues and facilitate dialogue that will lead to the formulation of practical, reasonable actions that assure the safety and wholesomeness of farm-raised products as they cross the producer's boundary and enter that of the processor on their way to consumers.

RESULTS & DISCUSSION

The Turning Years for Producers

Prior to 1989, the FDA Center for Veterinary Medicine (CVM) had little interaction with the U.S. aquaculture sector. During this era, aquaculture producers, researchers and others used a variety of drugs and chemicals with little knowledge of their legality or public health risks. Many drug and chemical therapy practices were based on "word of mouth" and

"commonly acceptable treatments" backed by empirical data or observations. Before 1990 there was a lack of accurate information on the proper use of legal drugs and chemicals. References available on the subject did contain errors which fostered an unquestionable use of unapproved drugs and chemicals among both the private and public sectors. However, in June 1989, CVM issued a high priority assignment to field offices to initiate inspections on private catfish, trout and crawfish farms. Eighty farm inspections were requested in this assignment. The objective was to determine which drugs/chemicals and feed additives were commonly used in aquaculture and the drug marketing strategies for these substances, and to obtain limited information concerning husbandry practices. Information on potential pesticide use was also recorded. At this same time CVM staff conducted a review of the literature to learn what information was available to producers regarding animal drugs and disease treatments.

The aquaculture sector was recognized as a new industry linked with an increasing per capita consumption of aquatic foods. CVM sought more comprehensive surveillance and compliance activities to keep pace with anticipated industry growth. The two substances of particular public health significance at this time were chloramphenicol and malachite green. Follow-up regulatory actions were directed at firms or individuals responsible for the promotion and distribution of illegal products rather than at individual aquaculture producers.

Initial FDA-CVM Surveillance Activities and Findings

A preliminary evaluation from this field inspection assignment revealed several concerns (11). There were various drugs and pesticides associated with multi-crop farms (that included aquaculture) and those solely producing aquaculture species that were not approved. Also, evidence indicated that chemicals might not be used in accordance with label instructions. There were no withdrawal times listed on the labels for some products marketed for use in aquatic animals.

Concern was raised that information about drugs and/or chemicals was not readily available to the aquaculture industry and current information was conflicting and/or not presented with the same emphasis as FDA desired. Much information lacked clarity on the liabilities incurred by producers who might use unapproved drugs in food animals. The suggested or recommended use of a wide variety of unapproved compounds in the scientific and educational literature indicated potential problems with the misuse of therapeutants by the industry. This was further substantiated by the concurrent field inspection assignment (11).

A FDA Committee was formed to conduct the study that focused on four objectives:

- identify current aquaculture production practices,
- review educational materials currently available and mechanisms for disseminating information,

- identify deficiencies in the information currently available to the industry and,
- develop recommendations for providing needed information.

Although these two surveillance activities were conducted independently the findings of each reinforced the other. The aquaculture literature review revealed the following (12):

- there is a lack of information on the drug approval process,
- a clear presentation of the approved drugs and their conditions of use had not been made to producers,
- limitations inherent in compounds approved for a specific use or condition have not been made clear and,
- compounds not approved for use in food fish, and with little data upon which to base such use, have been promoted for use in these species.

The on-farm field surveys found that unapproved compounds were present on most production facilities. These correlated well with those promoted in the literature. The presence of unapproved compounds on farms could imply their use in aquaculture production. This could potentially lead to the presence of violative residues in products going into food production. Recognizing the lack of reliable information and the wide availability of misinformation, CVM decided to focus initially on education as the primary means of addressing drug use concerns in aquaculture production (13). During this period, work also began on the development of new analytical methods to detect drug residues in aquatic species.

The FDA Committee developed the following recommendations:

- establish an aquaculture policy on proper drug use within CVM and FDA;
- establish a contact point for aquaculture information within CVM;
- coordinate the FDA aquaculture policy among its centers and offices;
- develop educational materials designed for specific target audiences and utilize existing avenues for distribution;
- develop liaison with industry and other professional organizations and;
- increase FDA-CVM visibility at aquaculture meetings, conferences and with other professional organizations.

There are also several matters unique to aquaculture that may have caused CVM to focus its regulatory attention more directly on the producer (14):

- the lack of a drug residue monitoring program similar to that for meat and poultry,
- the relatively few approved drugs for aquaculture and the resulting use of unapproved drugs by producers and,
- the use in aquaculture of general purpose chemicals that are not labeled for drug use, making regulation at the manufacturer/distribution level difficult.

Prior to 1990, numerous state extension programs emphasized avoiding the use of agricultural chemicals in the vicinity of commercial fishponds that could contaminate aquaculture crops, cause acute toxicities to fish or impact water quality through ground or aerial applications (15). There was also an increasing awareness concerning safety to workers when handling and applying aquacultural drugs and chemicals (16). At this time FDA-CVM had a liberal policy for issuing Investigational New Animal Drug (INAD) exemptions that was not well monitored or required the collection and submission of data from properly conducted clinical field trials to support New Animal Drug Applications (NADA).

CVM also concentrated on the manufacturers and distributors of drugs and chemicals that potentially could be used improperly by aquaculture producers (14). In June 1993 CVM issued an Aquaculture Plan. The Plan included a field assignment to inspect aquaculture drug manufacturers and distributors, specialty feed mills and commercial hatcheries during fiscal year (FY) 1993 (17).

Historical Background of HACCP Related to Aquaculture

As stated by FDA, "mandatory HACCP is a pioneering venture (8). Much of the pressure for a seafood inspection program was initiated by industry which considers a mandatory program a means to strengthen consumer confidence in the safety of seafood products and to become more competitive with meat and poultry. Consumer advocacy groups have also demanded Congressional action. During the period 1989-91, five separate congressional committees claimed jurisdiction over seafood safety and reported bills to overhaul current programs. However, there was no consensus on which federal agency should have regulatory jurisdiction, i.e., FDA, National Marine Fisheries Service (NMFS) or U.S. Department of Agriculture (USDA). With stalled legislation, NMFS and FDA agreed to move forward on a joint HACCP-based voluntary seafood inspection program based partly on a comprehensive model which NMFS had been researching since 1988 at Congress's request (18).

In 1992 FDA-Center for Food Safety and Applied Nutrition's (CFSAN) cooperative HACCP-based seafood inspection program efforts with NMFS transitioned into its own mandatory HACCP rules. While in mid-1992, NMFS encouraged seafood companies to

adopt a voluntary HACCP program through their new fee-for-service National Seafood Inspection Program (19). In January 1994 FDA issued a Federal Register notice on a "Proposal to Establish Procedures for the Safe Processing and Importing of Fish and Fishery Products" which was a HACCP-based system of preventive controls to ensure product safety (20). This proposed rule was not too surprising since a mandatory program had been in the making for over five years. After a period of public comment, FDA released its mandatory seafood inspection regulations on December 18, 1995. They become effective December 18, 1997, thus allowing industry 2 years to prepare for the implementation of the regulations (8).

The first exposure of the aquaculture community to HACCP occurred during 1989-90 when three Aquaculture HACCP Application Workshops were conducted in New Orleans, LA, Greenwood, MS and Seattle, WA. These workshops were organized by the National Fisheries Education Research Foundation of the National Fisheries Institute (NFI) with funding through a National Oceanic and Atmospheric Administration (NOAA)/NMFS Saltonstall-Kennedy grant. NFI in cooperation with NMFS was carrying out a Congressional mandate to design a seafood harvest-to-distribution surveillance program based on HACCP. The overall model program developed in this process was the basis for a recommended national seafood surveillance program that was presented to Congress for a decision on its implementation (21). Industry, academia, state agency and federal agency representatives provided information for the groundwork to develop the first HACCP Regulatory Model for Aquaculture under the Model Seafood Surveillance Project which was published and released by NMFS in 1991 (22).

In 1992 a video series was developed for application of HACCP in the catfish processing sector (23, 24). More recently two national satellite videoconference programs were broadcast to reach a national aquatic foods processing audience with pertinent information on HACCP compliance issues (25, 26).

FDA's Office of Seafood and Aquaculture Role

In 1990, the Administration requested and Congress approved a significant increase in FDA's FY 1991 seafood budget. Between FY 1990 and FY 1992, FDA's funding for seafood programs increased 80% to \$40.5 million. In February 1991 the Office of Seafood was created within FDA's CFSAN and in the same year CFSAN issued a Domestic Fish and Fishery Products Inspection Assignment (27). This project was aimed at Good Manufacturing Practice (GMP) inspections of all firms involved with processing fish and fishery products to determine the current status of fish and fishery processors in general and for use in future compliance programs/assignments. Collection and analyses of product samples were also included. Manufacturer and grower firms which process and package aquaculture stocks raised at that same facility were also inspected. Field inspections increased for both processors and producers and supportive research initiatives began with the creation of this Office.

FDA Office of Seafood 5-Year Strategic Plan

The Office of Seafood developed a 5-year strategic plan beginning in FY 1993 (28). Part of the Plan includes promoting and implementing the HACCP program throughout the seafood industry. In the U.S. this industry includes about 4,800 processors, 1,500 repackers and warehouses and 900 importers. This includes aquaculture products which are significant contributors to our overall seafood supply from domestic and imported sources .

The goal of the HACCP-based inspection system was to replace the current regulatory approach of spot inspections and end product sampling by the government with the industry monitoring itself on a continuous basis and the government verifying that the industry is implementing HACCP correctly. This involves inspections of CCPs and analysis of data demonstrating the processes are being performed within approved parameters. The strategic intent of the Office is to create complete consumer confidence in all seafood. The four basic goals of the strategic plan are:

- ensure that seafood products are safe,
- ensure that seafood is wholesome and is properly labelled,
- ensure that the FDA, the industry and consumers have accurate information concerning seafood and,
- ensure that the infrastructure exists to accomplish goals 1,2, and 3.

National Marine Fisheries Service Seafood Inspection Programs

NMFS original authority for seafood inspection dates back to the Agricultural Marketing Act of 1946 which was amended to transfer the development and promulgation of grade standards, the inspection and certification, and the improvement of transportation facilities and rates for fish and shellfish from USDA to the Department of Interior (DOI). In 1956, the DOI Fish and Wildlife Act established the Bureau of Commercial Fisheries (later to become NMFS) and its seafood inspection program. These inspection functions were transferred from DOI to the Department of Commerce (DOC) in 1970. Later in 1971 the Secretary of Commerce was charged with the administration of the regulations on Inspection and Certification of Establishments and Fishery Products for Human Consumption as amended in 1991 (29). In the 1970s NMFS issued guidelines to assist with the development of quality control systems for seafood processing operations. In 1975, followed by a revision in 1977, Federal Sanitation Standards for Fish Plants were developed (30). A continuous inspection service of monitoring and recordkeeping was first initiated followed by the Integrated Quality Assurance (IQA) program which was oriented to specified seafood products and based more on HACCP principles. In the 1980s federal standards for Good Manufacturing Practices (GMPs) were developed and plants had to meet new sanitary standards.

In July 1992 NMFS launched a new voluntary fish and seafood inspection service based on HACCP principles as a fee-for-service program and a refinement to their existing IQA

inspection service (19). Modifications to its voluntary seafood inspection program were made in 1993 based on several industry concerns with the existing program (31). NMFS began conducting HACCP training programs and certifying industry personnel as trained in HACCP principles and responsible for oversight of monitoring, recordkeeping and laboratory analyses. Under the new program NMFS conducts audits, end-product sampling and laboratory analyses to determine whether a participant's HACCP-based system is in compliance. NMFS will assist firms develop HACCP plans and will also evaluate plans as a consultative fee-based service. Products produced under the HACCP-based services may bear federal inspection marks.

These initiatives were influenced by a 1991 National Academy of Science report on microbiological seafood hazards which recommended the use of HACCP principles in the food processing industry (32). This same report recommended the application of HACCP at other points in the food chain, including production, storage, transport, retail sales and food service establishments. At this same time other countries were imposing HACCP-based inspection requirements, including the European Union (EU).

Numerous large-scale aquaculture processing companies enrolled in the NMFS program to "certify" their brand name products. Because of the fees required, small and medium processing companies generally have not participated and may experience some discrimination in the marketplace where "certification marks", which indicate that specific shipments have been federally inspected, may be expected or required. HACCP plans of firms must be approved by NMFS before firms can participate in the certification program.

NMFS continues to provide fee-based services to the seafood industry both domestically and internationally. The agency is also actively involved in seafood HACCP training and providing custom technical assistance upon request. NMFS is also recognized as a U.S. Government competent authority by the EU and signs health certificates as does FDA for seafood exports to EU member countries. NMFS was instrumental in developing the first HACCP Regulatory Model for Aquaculture under the Model Seafood Surveillance Project in 1991 (22).

FDA Focus on Baseline Pesticide Residue Monitoring

FDA had no data on the incidence of contaminant residues in major farm-raised species and was uncertain whether violative residues were a significant problem in aquaculture production similar to cases with other livestock production sectors. The data was also needed to determine whether regulatory enforcement actions would be needed. To add to the flurry of activity in 1990, that same year FDA initiated special surveys of aquacultural products under a specific program assignment (33). Work began in FY 1991 (27). The presence of pesticides and industrial chemicals in the aquatic environment was recognized as a potential for contamination of aquaculture food products. The objectives of the assignment were:

- sample and analyze domestic aquaculture products to obtain information on the incidence and levels of pesticides and industrial chemical residues and,
- initiate compliance sampling under the Pesticide and Industrial Chemicals in Domestic Foods Program for any products found to have illegal residues.

Investigational time was allocated to identify and locate aquaculture establishments. Primary target analyses were for organochlorine, organophosphorous and PCB residues. Priority sampled aquaculture products were catfish, crawfish, shrimp and shellfish with preference to bare earthen rearing ponds rather than lined or concrete systems.

In FY 1991, FDA issued a Domestic Fish and Fishery Products Inspection Assignment similar to the earlier CVM field assignment but different because products were sampled for residue screening (34). This field assignment was directed at seafood processors and aquaculture growers and included sample collections and testing. Specific background information was gathered through a rather detailed "Aquaculture Questionnaire" on production methods, species, substances used, medicated feeds, disease diagnostics, environmental conditions, transportation practices, etc. This work served as a basis for determining priorities for the development of residue detection methods for aquaculture products. In FY 1991, FDA initiated monitoring of imported aquaculture products for illegal drug or chemical residues.

Public Concerns and Issues with Residue Findings

When addressing issues associated with residues in foods, there are numerous sensitivities because of varying interpretations of the findings and public perceptions versus the reality of significant health risks. When a pesticide is found at trace levels in foods it is rarely a cause for public concern. From a regulatory standpoint trace levels of pesticides found in food are not considered to be of regulatory significance because these levels are found below the level of quantification and below a level that can actually be confirmed as a measurable amount. Science-based studies are required to determine that no human safety risk is associated with pesticide or drug levels that are below EPA established tolerances or FDA action levels. Tolerances are maximum legally acceptable levels for a specific use on a specific product that is set when a pesticide is registered. Action levels are triggers for regulatory action for residues of compounds for which there are no established tolerance. For example, chlorinated hydrocarbon pesticides have been banned for years but because of their long-term persistence in the environment, background levels in food crops can be found in areas where they were used in the past. Action levels have been determined when residues of numerous pesticides, that are no longer used, rise above acceptable limits associated with no human safety hazard.

Often the public does not understand the meaning of human health risk assessment or accept scientific studies that trace levels of pesticides are not harmful to their health. New generations of testing procedures and equipment can detect lower concentrations of

contaminants which can further raise public concern. Miscommunications can occur in the media about the risks of residues in foods because of the lack of facts or science. The aquaculture industry needs to identify potential safety risks and manage production effectively to avoid them. If a residue is detected that is not established for the specific crop or no tolerance has been set, then the food product is considered adulterated.

One problem with most pesticide residue databases and studies is that the data collected for fish and shellfish do not distinguish between farm-raised or wild harvested sources. Seafood residue problems and concerns are primarily associated with heavy metal or pesticide contamination in wild stocks of fish or shellfish where the environment is less controlled and managed compared to aquaculture production systems. There are still background levels of several pesticides that do exist in farm-raised species, but baseline data are within tolerances or action levels. Existing pesticide residue data for aquaculture products indicate an extremely low level of regulatory concern. However, the industry needs to continue to be aware of the importance of avoiding violative residues and follow vigilance and recommended practices concerning the proper use of drugs and chemicals.

Numerous processing plants routinely screen incoming raw product for pesticide residues to detect any potential problems. This however is limited primarily to the larger companies that can afford this expense. Smaller operations will rely more on producer adherence to QAPs and recordkeeping systems to assure that contaminants are prevented at the farm level.

Chemotherapeutant and Pesticide Residue Sampling Programs and Results

FDA has been active in sampling and monitoring pesticides and some drug residues in both domestic and imported aquaculture products for several years and is now reducing pesticide residue surveillance testing. FDA's Pesticides and Industrial Chemicals in Domestic Foods Program issued a field assignment for FY 1990 and 1991 for an aquaculture survey to determine the extent of contamination in aquaculture products from pesticides and industrial chemicals in the aquatic environment. (35). In FY 1990, a total of 172 samples was collected as follows: 103 catfish; 25 crawfish; 21 trout; 10 shrimp; and 13 other. These species were targeted because they represent the majority of aquaculture harvests. Although calculable and trace levels of several pesticides were detected, none of the residues found in this survey exceeded EPA tolerances or FDA action levels (36).

For FY 1991 FDA focused on persistent halogenated pesticides which may still be present in soils used for ponds as a result of past agricultural row crop use. A total of 188 samples was collected as follows: 128 catfish; 19 crawfish; 19 trout; 6 shrimp; 5 oysters and 11 other types. Again it was reported that no pesticide residues were found that exceeded EPA tolerances or FDA action levels (37).

FDA continued a drug and pesticide residue monitoring program specific for aquaculture products through FY 1992, 1993 and 1994. This program involved surveillance samples which are used to identify a potential problem. If a violative residue is found then the

appropriate FDA District Office is advised to conduct more vigorous official compliance sampling which can stand up to legal prosecution. Traceback investigations are done to identify the source of the problem and solve it. Beginning in 1992, surveys of selected aquaculture products were conducted under incidence/level monitoring. In FY 1992 a total of 206 samples included: 110 catfish; 43 trout and salmon; 32 crawfish and shrimp; 10 clams and mussels and 11 miscellaneous. A small number of samples contained trace levels of two chemicals with no tolerances. Follow-up actions reportedly revealed possible contamination from runoff or adjacent agricultural land use (38). No regulatory action was taken.

In FY 1993 a total of 308 domestic samples included: 121 catfish; 48 trout; 34 crawfish and shrimp; 18 salmon, 16 tilapia and 40 other fish and shellfish. Findings revealed a few isolated cases of trace levels of three chemicals with no tolerances but follow-up steps included no regulatory action (39). In the FY 1994 survey, FDA collected 160 samples including: 39 catfish; 31 crawfish and shrimp; 30 trout; 22 salmon; 16 oysters and 22 various other products. Again, a few chemicals were found in isolated samples, but no persistent halogenated pesticides were found at levels that exceeded FDA action levels (40). In all sampling work from 1992 to 1994 there were no organochlorine or organophosphate pesticides found in the aquaculture products sampled that exceeded FDA action levels. Several years ago EPA eliminated the tolerance for toxaphene in fish because background levels in the environment have for the most part become insignificant.

After FY 1994 FDA curtailed its domestic aquaculture pesticide residue monitoring program that specifically identified aquaculture products that first began in 1990. This decision was based on dwindling resources and the fact that no significant safety problems or concerns were identified. Aquaculture products continue to be included in FDA's pesticide monitoring program but not as specifically coded products. Farm-raised products are included with other sources of seafood under minimal guidelines and pesticide residue findings are reported generically for fish and shellfish and not by species or source. It is now difficult to extract data for aquaculture species. Data from the FY 1995 pesticide residue monitoring program do not distinguish aquaculture products from other sources of fish and shellfish, but no violative residues were reported from a total of 295 fish and 90 shellfish sampled (41). Pesticide residue findings from the FY 1996 survey have not yet been published.

In other action, FDA-CFSAN began routinely sampling and testing for various residues in aquaculture products in FY 1992 under several field assignments which are ongoing (42). The field assignments for FY 1992-94 addressed sampling of both imported and domestic shrimp for residue levels of chloramphenicol. For FY 1994-95 the assignment was extended to oxolinic acid in imported and domestic salmon after the analytical detection method was developed. In 1996, the compliance program assignments were combined into a mechanism to detect violative residues. The basis for this program includes the fact that imported food accounts for an increasing portion of total U.S. seafood consumption and there is an increasing concern over unapproved drugs and other chemical agents used to prevent and/or control aquatic weed growth. FDA seeks to obtain data to develop a clear picture of the

current levels of antibiotic residues in farm-raised aquatic food products. Regulatory action is initiated when warranted.

Preliminary data suggest that of the domestic species collected and analyzed for a limited number of drug residues no significant antibiotic residue problems exist (42). Additionally, in the first on-farm field inspection assignment conducted by CFSAN in FY 1991 for domestic aquaculture growers, which included residue screening of samples, no residues were reportedly found according to FDA officials. However, in FY 1992, 3 out of 50 shipments of imported farm-raised shrimp sampled contained chloramphenicol and were refused entry (43). For FY 1993, 2 out of 100 samples of imported farm-raised shrimp tested positive for chloramphenicol while no imported farm-raised salmon tested contained drug residues (Kim Young, FDA, personal communication). However in FY 1994, 1 out of 43 samples of imported farm-raised salmon tested positive for oxolinic acid, and 1 out of 54 samples of imported cultured shrimp tested positive for chloramphenicol (Kim Young, FDA, personal communication). For both FY 1995 and 1996 no violative residues of chloramphenicol in imported farm-raised shrimp or oxolinic acid in imported farm-raised salmon were detected out of 36 shrimp and 75 salmon samples, and 45 shrimp and 55 salmon samples, respectively (Kim Young, FDA, personal communication). More compounds can be expected to be included in this ongoing drug screening compliance program in the future as detection methods are developed for other drugs for various farm-raised species. If a residue problem is found at a processing facility, FDA through CVM then traces fish samples back to a specific farm to identify the source of contamination and institutes needed remedial or regulatory action.

Another effort that has generated baseline information on the incidence and levels of residues in various farm-raised species is a 3-year project initiated in 1992 with funding by the USDA/Cooperative State Research, Education and Extension Service (CSREES) through the Southern Regional Aquaculture Center. The overall goal of this project was to assure the quality and safety of aquacultural products reaching consumers. The testing program for residues was conducted to determine any real or potential problems regarding the safety of southern aquacultural products. This project involved development of a database for chemical contamination in farm-raised channel catfish, crawfish and rainbow trout, formulation of guidelines and protocols for a residue monitoring program at a processing facility, development of educational materials for producers and processors concerning the safe use of chemicals, adaption and dissemination of existing chemical application recordkeeping systems for aquaculture producers, determining the fate of residues from the farm to processing plants, and conducting additional sampling of aquaculture products to improve the database. The residue sampling and testing work involved numerous organochlorine and organophosphate pesticides, heavy metals and several pyrethroids (44). Results of the residue sampling work from this extensive project have not yet been published but the preliminary findings have indicated no significant residue problems in the product samples tested (George Lewis, University of Georgia, personal communication).

Development of Analytical Detection Methods for Regulatory Enforcement

When FDA first began to develop regulatory plans for the aquaculture sector, including both domestic production and imports, the agency lacked approved drug analytical detection methods that could withstand legal scrutiny in court. The FDA directed resources for drug methods development for assaying residues of unapproved drugs in aquaculture species. These analytical methods are needed regulatory tools and are linked to compounds of the highest human safety concern. Method development research to detect drug residues in different aquaculture species began in 1992 and continues today (Table 1). FDA research is focused on hazards posed by microbial pathogens, chemical and drug residues, marine toxins, parasites, decomposition and new packaging technology.

Table 1. Aquaculture drugs with residue detection methods completed or planned for Compliance Program use.

<u>Drug</u>	<u>Species</u>	<u>Compliance Program</u>	<u>Status</u>
Chloramphenicol	Shrimp	Domestic/imports	Implemented
Oxolinic acid	Salmon	Domestic/imports	Implemented
Malachite green	Catfish	Domestic	Implemented
Oxytetracycline	Shrimp	Domestic/imports	On hold

In addition to work in progress, FDA has already identified a priority listing of aquaculture drugs and chemicals for future methods development work. The pace of this activity will depend on budget, laboratory and personnel resources. FDA is completing construction of an aquatics wet laboratory that will provide additional facilities to conduct intramural aquaculture drug/chemical residue methods development research beginning in 1997. FDA will also have the capability to conduct research on drug metabolism to better understand residue depletion rates for individual drugs in various aquatic species and at different temperatures. (Norris Alderman, FDA-CVM, personal communication).

Role of Federal Joint Subcommittee on Aquaculture

The Federal Joint Subcommittee on Aquaculture (JSA) functions as a Subcommittee of the National Science and Technology Council's Committee on Health, Safety and Food. The JSA is chaired by a designate of the Secretary of Agriculture and joint leadership is provided through an Executive Committee with representatives from USDA, DOC and DOI. The JSA serves as a federal interagency coordinating body and forms national task forces or working groups as needed to address issues of national scope and importance.

In 1990 the preliminary FDA-CVM field surveillance findings were presented to the JSA. Because of the implications of food safety and the national scope of the findings, the JSA established a forum for public and private sector dialogue to engage the U.S. aquaculture community to address and resolve concerns and priority issues. In November 1990, an interagency coordinating group called the Working Group on Quality Assurance in Aquaculture Production (WGQAAP or abbreviated WG) met formally to initiate strategies to address FDA's concerns. The WG now has more than 100 participants, including

representatives from Canada. Industry representatives also serve as WG participants to create the needed public-private sector national forum to identify priority issues and develop resolution strategies and actions (45).

This WG has accomplished significant change and impact in the aquaculture sector. Any recommendation or request for action to any federal regulatory agency is not associated with the WG but is initiated by the independent actions of trade organizations, commodity associations, professional societies or individuals. This arrangement keeps WG meetings in compliance with provisions of the Federal Advisory Committee Act and avoids potential conflicts of interest.

Accomplishments of the JSA Working Group

Since its inception, the WG has been effective in assembling key public and private sector stakeholders associated with aquaculture. An important aspect is that FDA fully participates in the WG and collaborates on educational initiatives with its members. The WG directed immediate attention to the education of producers. Support came for an aquaculture drug/pesticide database as a component of an existing USDA-funded Food Animal Residue Avoidance Databank (FARAD) project. This database now monitors product label information and scientific literature on drugs, chemicals and pesticides that can be used in aquatic sites and aquaculture. The program also provides guidance to veterinarians when prescribing extra-label use of drugs. A software program was developed recently to make database information available to the public and is presently being evaluated for its utility (46).

The WG has been instrumental in developing several educational products (47,48), supporting producer QAPs, sponsoring workshops and special sessions at international, national and regional conferences, identifying drugs of highest priority for approval, creating a National Aquaculture New Animal Drug Application Coordinator position through public and private sector funding, providing leadership for international harmonization efforts for aquaculture drugs and biologics, and serving as a credible national forum for public discussion of aquaculture drug issues.

Efforts of the WG, linked with the development of educational literature and proactivity by producer associations through their QAPs, have increased the awareness of producers on the critical importance of proper drug and chemical use to avoid violative residues. Coordinated work focused on new animal drug approvals for aquaculture species has resulted in strategic public-private sector partnerships and good progress in developing data to support the FDA approval of needed new animal drugs. Additionally, research and on-farm experience with traditional therapeutic treatments against common disease outbreaks have resulted in new management practices to minimize losses from disease pathogens and/or let the disease run its course without drug treatment therapy.

The WG recently completed a new 5-year strategic plan to continue this collaborative structure into the future as needs warrant. More emphasis is expected on biologics. One new challenge is stronger interaction with the processing sector, as mandatory HACCP regulations link pre-harvest practices to potential food safety hazards which need to be addressed.

Producer-Quality Assurance Programs (QAPs)

When processors receive raw incoming product for processing, they assume responsibility for the condition of the product, regardless of how producers and others are regulated. With new mandatory food safety regulations aimed at the seafood, meat and poultry sectors, food animal producers are going to be impacted by this transition into an expanding farm to fork HACCP food safety spectrum. Processors will be seeking assurances that the raw products they receive are safe from contaminants. Producer implementation of commodity food safety and QAP practices will become increasingly important to processors and consumers. Industry is ultimately responsible for producing and marketing high quality, wholesome products. HACCP concepts in residue avoidance should be incorporated into all educational quality assurance and good production practice programs (49).

The voluntary QAPs provide producers with the guidance that will allow them to address processor concerns about violative residues. The USDA Food Safety and Inspection Service's (FSIS) mandatory food safety regulation for meat and poultry processing plants includes the strategy for the voluntary use of food safety and QAPs, based on HACCP principles, to reduce pre-harvest food safety risks (50). Implementation of QAPs can increase consumer confidence and the value of marketable products in addition to improving production efficiency. Emphasis on good husbandry practices can in fact prevent and/or reduce disease and decrease the need for therapeutants.

In the catfish and trout industries a small number of large processors market the majority of the production volume. Their acceptance of QAPs and decisions on HACCP protocols for pre-harvest safety concerns have significant implications industry-wide. Most, if not all, are already familiar with HACCP and operate under a continuous government inspection certification program offered by NMFS.

Producers need to demonstrate that they have used drugs, chemicals and pesticides properly and according to industry and regulatory standards. Continued public concern about residues and foodborne illnesses is an important incentive to food animal production industries to implement HACCP-based systems, even if they are not required. The mandatory pressures on processors and importers will likely have a ripple effect on producers because more information about on-farm practices may be needed from producers to satisfy the safety performance measurements of processor HACCP plans. Producer implementation of QAP practices can reduce the need for verification testing and allay concerns about contamination that can occur during production. These practices can assist processors in meeting much of

their pre-harvest residue control responsibilities if a CCP is identified with receiving raw cultured products..

FDA-CVM began working with traditional animal agriculture commodity groups to promote QAPs and encouraged their development by different industry sectors (51). FDA-CVM has stated that voluntary industry QAPs which incorporate recordkeeping procedures to manage husbandry practices and control drug use, can provide a producer with a mechanism to demonstrate that proper controls are in place (52). FDA advocates that the best avenue for producers to ensure and document that drugs are used properly is through quality control or QAPs (53). FDA also notes that while QAPs have been established they will not be effective in improving the overall food safety of aquaculture products unless there is industry-wide participation and a sincere commitment to their principles (53). The clear message is that promoting proper animal drug use practices requires the cooperation of everyone involved in food animal production. This is spurred by growing consumer awareness and concerns about potential health hazards and perceptions associated with drug, chemical and pesticide uses in agriculture animal production.

The existing QAPs relate differently to HACCP principles and are for the most part based on good management practices. Producer enrollment is variable and further efforts are needed to educate producers on how these programs can improve the industry and on-farm operations. Food safety is a public health issue with significant economic and social costs and the public needs to understand the safeguards involved in aquatic food production as a segment of the total food system, participating as equal partners in pathogen and residue avoidance and reduction. Processors will be responsible for safety assurances and justifiably will monitor the safety of the farm-raised products entering their plants as part of their HACCP program - their first CCP.

A new approach, called Total Quality Management (TQM), integrates HACCP and contains most of the elements needed for an on-farm quality and safety program. TQM requires that management focuses on quality and safety and develops standardized approaches and records, and monitors to deliver desired outcomes. Its usefulness as a management tool for producers becomes more apparent with new regulatory changes and mandated HACCP in processing plants. Farms can control as well as document production practices, sensitive to the needs of their customers. This sense of understanding and control is the foundation of TQM. QAPs and TQM can assist producers to develop on-farm systems to complement a regulated HACCP program imposed on processors by the government. However, support materials, expertise and training initiatives are needed to move in this direction. QAPs can be an effective vehicle and foundation for any TQM transition.

QAP Generic Resources and Educational Initiatives

Significant strides have occurred in the development of producer QAPs within the aquaculture sector. The issue began receiving serious attention by the WG in 1991-92 with various strategies proposed (54). The heightened awareness of the need for QAPs generated

numerous projects and products to educate producers nationwide. The industry took proactive actions in collaboration with state Extension Service programs to develop various generic aquaculture QAP educational products (55) and broadcast several national satellite videoconferences (56,57). A special forum was organized to address the issues of microbes and residues and their relationship to the safety of Southeastern U.S. aquacultural products (58). To apprise the U.S. aquaculture industry of available materials, a bibliography of resources on HACCP and QAPs was developed and distributed widely on the Internet through mailgroups and home pages in addition to an industry newsletter (59). USDA-CSREES provided funding support for development of QAP-related educational products and implementation of numerous educational initiatives linked to support the awareness of QAPs among producers nationwide through the Cooperative Extension system and Sea Grant Marine Advisory Service (60).

Catfish Industry Leadership. The concept of QAPs was supported by several national aquaculture associations. The Catfish Farmers of America (CFA)(5) developed the first program in 1993 in cooperation with state Cooperative Extension Services in the southern region based on a needs assessment (61). The effort also included participation of The Catfish Institute - the marketing arm of the industry, and recognized the importance of QAPs to all aquaculture industries as a whole. Much of the processing sector was already implementing QAP and was enrolled in the NMFS voluntary HACCP inspection program for processors. Partial funding to print the CFA-QAP was contributed by The Aquaculture Council of the National Fisheries Institute. The program is "an educational program designed to maintain the consumer love affair with farm-raised catfish." The program addresses potential on-farm human food safety control points but is not considered to be a HACCP plan for producers. A Voluntary Enrollment Card is included to acknowledge a review and understanding of the program. The program is promoted by industry leaders and Extension specialists and at national conferences. More work is needed to increase total production associated with the program which is currently about 45% of the total catfish acreage. The catfish QAP was reprinted with some updates in 1997.

Trout Industry Initiative. The trout industry was first introduced to QAPs at an annual conference in 1991. In 1994 the U.S. Trout Farmers Association published a Trout Producer Quality Assurance Program (6). It is "an educational program designed to ensure continued production of high-quality, wholesome farm-raised trout." Two parts comprise the program. Part 2 is based on HACCP principles and identification of potential CCPs. It involves development of a producer HACCP plan, an effective recordkeeping system and verification controls. The program also includes a producer Pledge and Registration Card but takes matters one step further with a Verifier's Affidavit. This affidavit is a statement by an outside third party that the producer has a QAP with all safety and quality controls being properly implemented and recorded correctly. The program was developed by a coalition of industry leaders and Cooperative Extension specialists from several trout producing states. Currently,

about 50% of trout foodfish production or about 25 million pounds annually are covered in this QAP.

Other Aquaculture Producer Directed Programs. The hybrid striped bass sector recently completed a publication that includes elements of a QAP with a HACCP Model for Aquaculture (62). The project was funded by the USDA Agricultural Marketing Service. Two generic QAPs for finfish and shellfish producers are being developed by the National Aquaculture Association with funding support from USDA's Northeastern Regional Aquaculture Center (63,64). Several salmon companies have installed HACCP plans for their farms and plan to use that safety measure in marketing their products (65).

The Relationship Between FDA Compliance Policy Guide and Quality Assurance Programs

FDA-CVM is responsible for regulating and enforcing matters pertaining to the approval, proper use and residue violations of animal drugs in food-producing animals. In 1993 the agency issued a Compliance Policy Guide which addressed proper drug use and residue avoidance by non-veterinarians (66). If QAPs adequately address the policy component pertaining to avoiding drug residues through proper drug use, then QAP control systems and programs should be recognized and accepted in processor HACCP plans. This should also offer a stronger case for the acceptance of supplier guarantees or certificates.

The control systems encouraged in this policy guide are for persons involved in raising, handling, transporting, holding, and marketing food-producing animals to ensure animal drugs are used properly and to prevent potentially hazardous drug residues in edible animal products which include the following measures:

- Identifying and tracking animals to which drugs were administered;
- Maintaining a system of medication/treatment records that at a minimum identifies the animal(s) treated, the date(s) of treatment, the drug(s) administered, who administered the drug(s), the amounts administered, and the withdrawal times prior to slaughter;
- Properly storing, labeling, and accounting of all drug products and medicated feeds;
- Obtaining and using veterinary prescription drugs only through a licensed veterinarian based on a valid veterinarian/client/patient relationship; and
- Educating all employees and family members involved in treating, hauling and selling the animals on proper administration techniques, observance of withdrawal times, and methods to avoid marketing adulterated products for human food (66).

Establishing and maintaining such systems should help producers avoid marketing products containing illegal residues and avoid regulatory action. All QAPs should adequately address these control systems and follow FDA recommendations. In FDA's view, failure to maintain adequate controls with respect to the use of animal drugs could result in a reasonable possibility of injury to human health because illegal drug residues often result from a lack of such controls, and illegal drug residues could have adverse toxicological effect on consumers, ranging from acute to chronic reactions (66).

The use of animal drugs in aquaculture production is subject to the requirements in the Federal Food Drug and Cosmetic Act (FFDCA). FDA concerns about aquaculture producers' use of drugs are heightened because of several unique circumstances which include:

- The lack of a drug residue monitoring program similar to that for meat and poultry;
- The relatively few approved drugs for aquaculture and the resulting pressure for producers to use unapproved drugs; and
- The use of aquaculture general purpose chemicals that are not labeled for drug use, making regulation at the manufacturer/distributor levels difficult (67).

FDA's enforcement focus has been on drug manufacturers and distributors that produce and distribute drugs for aquaculture use. The goal has been to cut illegal drugs off at their sources (53). Medicated feed manufacturers have been the next level of regulatory priority. CVM's emphasis has been on education on proper drug usage and how to comply with HACCP (53). CVM will continue to consider enforcement action against violative drug residues to be a high priority. The focus on education was in response to the situation in past years.

FDA Response to Supplier Guarantees

In the seafood HACCP regulations (8), several processors recommended that a certificate from a producer indicating that a lot of raw fish material is free from unacceptable levels of pesticide and drug residues should be an acceptable means of monitoring the hazards of animal drug and pesticide residues in aquaculture-raised fish. Comments also stated that the certificates should be based on participation in an industry-wide QAP designed to ensure that the raw materials are free from these hazards. These comments are appropriate and what has been advocated by industry and strongly supported by FDA-CVM (52). However, in the seafood HACCP regulations, FDA-CFSAN responded that caution is warranted on this subject and more direct controls should be used if available, which FDA contends may include a review of supplier's animal drug control records and a system of on-site audits of the supplier either by the processor or a third party. FDA also recognizes cases where such controls are not possible, and suppliers' certificates or guarantees are the only available monitoring tool. The critical issue to this approach is the verification of the effectiveness of the certificates. FDA also commented that the extent to which suppliers' guarantees can be relied upon will have to be considered on a case-by-case basis.

These responses and the position of FDA regarding the integration and use of QAPs to address contamination concerns through producer participation in QAPs or through supplier guarantees or certificates need further clarification. Producers and processors should proactively seek reasonable means to address human safety risks using existing available programs and services. There is also some conflict regarding earlier guidance by FDA-CVM on QAPs meeting HACCP requirements. An adequate recordkeeping system and reporting by producers should provide the bridge between producer QAPs and processor HACCP plan requirements regarding potential safety risks from contaminant residues. With credible and effective producer QAPs, including proper verification protocols, the producer certificate or guarantee should be another reasonable option.

Some QAPs may require modifications or updating to match-up closer with the HACCP process. One issue of particular concern to FDA is verification that producers are implementing these programs as intended. Some animal agriculture industries are revising their QAPs to incorporate HACCP procedures in addition to best management, pre-harvest practices required for high product quality and safety (68,69). The industry QAPs provide the guidance to producers regarding what they need to do to meet food safety and regulatory standards. Producers must implement these practices. It is only when they actually implement them that their responsibility is met.

Seafood HACCP Regulations Definitions

There are several issues that are germane to those involved with aquaculture products that relate to definitions. For example, there are numerous situations when producers or paylake operators custom process fish on-site for their customers. This is a value-added service offered after the fish have been purchased by the public. Is this regarded as a retail establishment and therefore exempt from the regulations? In the seafood HACCP regulations the following definitions should be understood because they have implications based on their interpretation for the different segments and practices in aquaculture (70):

Fish means fresh or saltwater finfish, crustaceans, other forms of aquatic life {including, but not limited to, alligator, frog, aquatic turtle, jellyfish, sea cucumber, and sea urchin} other than birds or mammals, and all mollusks, where such animal life is intended for human consumption.

Fishery product means any edible human food product in which fish is a characterizing ingredient.

Processing means handling, storing, preparing, heading, eviscerating, shucking, freezing, changing into different market forms, manufacturing, preserving, packing, labeling, dockside unloading or holding. The regulations in this part do not apply to:

- (i) harvesting or transporting fish or fishery products, without otherwise engaging in processing,

- (ii) practices such as heading, eviscerating or freezing intended solely to prepare a fish for holding on a harvest vessel and,
- (iii) the operation of a retail establishment.

Processor means any person engaged in commercial, custom, or institutional processing of fish or fishery products, either in the United States or in a foreign country. A processor includes any person engaged in the production of foods that are to be used in market or consumer tests.

Food safety hazard means any biological, chemical, or physical property that may cause a food to be unsafe for human consumption.

Critical control point means a point, step or procedure in a food process at which control can be applied, and a food safety hazard can as a result be prevented, eliminated, or reduced to acceptable levels.

National Seafood HACCP Alliance for Education and Training

In the seafood HACCP inspection regulations, FDA requires that all processors either employ at least one trained individual or contract services from at least one trained individual, as needed. FDA has been extensively involved with the National Seafood HACCP Alliance to create a uniform core training program that will meet the requirements of these regulations and will cost very little (8). FDA also allows job experience to serve as a form of training as long as it is equivalent to that provided by the course. This Alliance was formed in 1993 as a cooperative effort involving federal and state agencies, academia and industry trade organizations with an interest in developing an educational framework to assist the seafood processing sector in complying with the anticipated mandatory HACCP-based seafood regulation. The National Sea Grant College Program provided a grant to develop a HACCP training curriculum which was revised in 1996 (70). The latest revision of the curriculum does recognize the utility of producer QAPs as a means of addressing and preventing potential pre-harvest human safety risks for contaminants.

This curriculum is important because it is a collaborative public-private sector effort with the involvement of FDA and NMFS in developing a standardized training curriculum for use by regulators and industry. A train-the-trainers program was initiated through the regional affiliates of the Association of Food and Drug Officials (AFDO). This effort has resulted in over 400 AFDO certified seafood HACCP trainers who can organize training programs for industry and other interested parties and use the Alliance's curriculum as an educational resource. Courses are being offered throughout the U.S. A Compendium of processing methods and techniques has also been developed as a resource for processors in addition to the identification of research priorities associated with the implementation of HACCP in the seafood sector.

Participants in the Alliance have also been instrumental in developing a variety of HACCP implementation manuals, through USDA-CSREES funding, for specific processing operations of different aquaculture species. These species include catfish, trout, crawfish, oysters, hard clams, and mussels (71,72,73,74,75,76).

International Trading Pressures

More countries are recognizing HACCP in food safety legislation and consider HACCP as a great potential to facilitate international trade. Demands for stricter standards from importing countries are also increasing. For the U.S. to compete in international markets there is a growing trend toward documenting safety and quality processes for other countries, including member countries of the EU, Canada and others. Presently the value of exports for domestic aquaculture food products is estimated at \$25-30 million annually depending on foreign currency exchanges and market conditions (David Harvey, USDA Economic Research Service, personal communication). The U.S. is involved with equivalency agreements with other countries that include fairness and consistency, transparency of process, no loosening of standards and proper verification. FDA is concerned with an appropriate level of consumer protection.

The Food and Agricultural Organization (FAO) Codex Alimentarius Committee has 154 member countries and represents 97% of the world's population. The Codex Committee on Fish and Fishery Products has developed a Proposed Draft Code of Hygienic Practice for the Products of Aquaculture (77). This Code is being drafted by the FAO and World Health Organization (WHO) and includes HACCP principles and a model plan as guidance to fish farmers. Codex is filling a niche that no single country can accomplish and its actions will likely have a profound impact on improved global consumer protection. The issues of equivalent systems and recognition of alternative food safety systems may represent trade barriers. These issues have caused the Organization for Economic Cooperation and Development's (OECD) Committee for Fisheries to assess incompatible seafood inspection standards and requirements that often cause trade disruptions and financial losses for seafood exporters and higher prices for consumers.

Other international organizations involved in HACCP-related issues for seafoods include the Sanitary and Phytosanitary Standards (SPS) under the World Trade Organization (WTO) and the International Office of Epizootics (OIE). The concern is that HACCP will become the global standard for seafood inspection in the future. The major problem to a successful determination of equivalence of different systems is the varying degree in the HACCP system's application. The basic definition of equivalency is the capability of different inspection and certification systems to meet the same safety objectives.

The EU is harmonizing a HACCP-based inspection program to be implemented in the seafood processing sector for all 15 member countries. In 1991 the EU (formerly known as the European Community) issued a series of regulations or Directives on Fishery Products including specifically Directive (91/493) which placed new responsibilities on the U.S.

government and seafood exporters. One element is the requirement that U.S. exporters to EU countries be included on a listing of approved establishments which have been determined by a competent authority (FDA or NMFS) to have controls and conditions equivalent to the requirements of the EU (78).

In March, 1993 the EU issued Decision 93/185 which provided a transition period for fishery products and raw molluscan shellfish. This period was provided so that EU and third party countries could harmonize the conditions under which these products are regulated by implementing equivalent or HACCP type control measures. The transition period for this new requirement began on July 1, 1993 and ended December 31, 1994. During this time the EU required that each entry of fishery products be accompanied by an EU "health certificate" signed by a U.S. government competent authority. Presently, FDA maintains a national inventory of fishery product firms each with an identification number for tracking all agency activities associated with any firm. Any U.S. company that desires to export seafoods to EU member countries needs to be officially included on the U.S. Government's list of acceptable firms for having a HACCP program in operation that is equivalent to EU requirements.

Other countries, for example those in Latin America, have signed Agreements with the EU based on principles of equivalence and include mutually agreed inspections by EU officers to Government facilities and the industry. Most of the new Latin American seafood regulations are based on the HACCP Guidelines of the FAO/WHO Codex Alimentarius Committee which in turn are the international legal reference on this matter with the General Agreement on Tariffs and Trade (Uruguay Round) Agreements.

CONCLUSIONS

Even though new food safety regulations are targeted directly at seafood processors and importers, the requirements will likely have an indirect effect on aquaculture production and marketing. The seafood sector will not only be influenced by its mandatory inspection program but also that of other animal protein industries - meat and poultry. Consumers and the public will become more aware of HACCP as a food safety program and its link from farm to fork. The growing sense is that those involved in each segment of the farm to table food process, from producers to retailers and consumers, bear responsibility for identifying and preventing or reducing food safety hazards that are under their operational control (50).

FDA has directed considerable resources to incorporate aquaculture production and seafood processing into existing and new surveillance and regulatory programs in the last several years. The emphasis has been on education and gathering information from producers and processors to evaluate potential safety hazards associated with this food sector. Pesticide and drug residue screening of domestic aquaculture products has not revealed any human safety problems or violations that have required regulatory action. However, this involves only a few compounds and FDA will continue drug-pesticide residue monitoring and

increase product sampling for other drug residues when new analytical detection methods are validated.

Progress on development of new animal drug approvals is continuing; however the process is protracted and costly. Through the cooperation and diligent efforts of many public and private aquaculturists, significant pivotal and supporting data are being generated toward needed new animal drug approvals. New approaches with crop grouping to obtain approval for multiple species rather than individual species and definitions of early life stages that are treated exclusively with a certain product are expected to provide some relief. FDA's recent approval of formalin for fish generically, rather than for individual species, is an indication of significant progress. Outreach into the international arena to foster multi-country collaboration on drug and biologics approvals for aquaculture and establish maximum residue limits (MRLs) for drugs of importance for international trade is another long-term initiative that requires an advocacy for aquaculture among international organizations and communities.

Progress in the development of QAPs has been quite remarkable over a short span of time and new initiatives are still under development. The challenge is to highlight the value and need for such programs among producers. These programs enable safety to be an element in every critical step in the production process, including an accurate disease diagnosis, selection of the most appropriate mitigation (management, prevention or therapy), recordkeeping systems and review of treatment information as on-farm residue avoidance procedures prior to the sale of aquaculture products. This approach is far superior to end product testing. These programs can benefit the economics of production and science-based best management practices can become important industry guidelines.

Although the domestic aquaculture sector has a good record of contaminant avoidance, the potential risk is ever-present and proactive preventive measures are always needed. One significant incident broadcast widely in the media could taint the public's image of the safety and quality associated with aquaculture or farm-raised food products. The public's perception is with the product and its related production practices. It is in the best interest of the entire U.S. aquaculture sector to embrace QAPs and be proactive partners in the farm to fork HACCP spectrum. QAPs focus on prevention and avoidance of residue problems and can ease requirements for both processors and producers. It is essential that all producers recognize that they have a critical stake in maintaining consumer confidence in the safety and quality of farm-raised products. This effort involves the entire sector from feed manufacturers and speciality product suppliers to producers and processors.

The advent of HACCP after much discussion for many years is now approaching. The processing sector and importers of seafood products are currently reacting to the seafood HACCP inspection regulations and effective date. Many larger processing operations have been operating under HACCP with in-plant inspections supervised by NMFS. However, smaller processors have not been impacted until now and the expectations from producers

to satisfy any processor's HACCP plan remains uncertain for many. Producer and processor responsibilities for aquaculture food safety are now converging.

The FDA guide (10) cites Chapters on Environmental Chemical Contaminants & Pesticides and Aquaculture Drugs. For each potential chemical hazard there are two critical issues. First an understanding of the potential hazard and secondly a determination if this potential hazard is significant. These are fundamental issues and the outcomes may vary among individual processors and the interpretation of significance may also vary among individual government plant inspectors. QAPs provide the means for processors to better evaluate the significance of these potential hazards as they are addressed in QAPs as preventive measures. A knowledge of existing data and development of new data may also contribute to an assessment of human safety risks.

Present residue monitoring data reinforce the fact that the incidence of residue problems is extremely low. However, this evaluation is based on non-statistical screening surveys and few compounds. Pre-harvest safety risks from domestic farm-raised finfish should be regarded as "low" and addressed in a reasonable manner by producers and processors in the HACCP process. Hence, aquaculture producers and processors should collectively address these issues now with FDA consultation to clarify satisfactory options for addressing these potential risks in processor HACCP plans. If the decision is made that these are significant risks then another level of requirements for producers and processors will result with identification of CCPs, setting critical limits and adopting control strategies that can involve sampling, testing and verification. The language in the Guide (First Edition) is important because FDA considers a comparison between a processor's HACCP plan and the Guide to be a level of verification.

Some processors have established safety criteria for producers and others have actually developed Certificates of Compliance for producers to sign that reference the application of the Catfish Quality Assurance Program. This cross-referencing and recognition of QAPs by processors will further enhance adoption of these programs nationwide by producers of all species. However, these programs lack a tested verification component. CVM and FSIS are encouraging industry commodity organizations to develop their own QAPs. These voluntary programs can assist producers in implementing proper drug and pesticide use practices on farms and recording data when using an investigational drug as a clinical investigator (79).

Producer QAPs can facilitate compliance with recordkeeping requirements and prevention of environmental contamination of farm-raised products. The programs are educational but do require a commitment to HACCP principles with some gaps, especially in acceptable verification protocols. Processors are in a position to demand producer participation in QAPs or similar programs to protect themselves as they are responsible for assuring product safety through their HACCP plans. This will only work if producers and processors jointly develop fair, yet adequate safety controls and reporting measures and each realizes their role in maintaining consumer confidence with high quality, safe products.

While one FDA center (CVM) promotes QAPs as a means of complying with the pre-harvest recordkeeping requirements linked to the processors' mandatory seafood inspection regulation, another FDA center, CFSAN, which developed the seafood HACCP inspection regulations, did not mention nor endorse QAPs in these regulations (8) or in the Guide (10). This oversight was unfortunate because the two programs - one voluntary and the other mandatory are complementary and serve as critical linkages between producers and processors. Both sectors are stakeholders and stewards to assuring the safety and quality of farm-raised products that reach consumers wherever their location. The Guide was the agency's initial effort and the next version will include references to QAPs and their role in HACCP plans (Kim Young, FDA, personal communication). However, because of the lack of adequate verification protocols, QAPs were not considered to be an adequate safety program by FDA-CFSAN when these documents were developed. This is an issue that producers will have to address or clarify.

Many processors are going to consider the incoming raw products to be a CCP because of the need to ensure adequate control measures of illegal chemical residues. FDA's Guide and language in the seafood HACCP inspection regulations imply that a potential CCP, in fact the first CCP, may be receiving raw product sourced from aquaculture producers. Concerns include violative drug residues, use of illegal therapeutants, inadequate withdrawal times after treatment, contamination from environmental hazards such as pesticides, heavy metals or other compounds of concern to human health and possible microbial contamination from production environments or product handling. This puts processors in the position to fully understand and adopt adequate control measures to avoid violative residues. A hazard analysis could include any potential safety hazard including feed additives, drugs, chemicals, pesticides or other contaminants that incoming product may be exposed to. Processors may need sound evidence of producers' efforts to prevent violative residues (49).

Producers are responsible for supplying processors and consumers with high quality, safe, wholesome products. This includes all production, transportation and handling of product until it reaches the processor's dock or holding tanks. Because consumers associate farm-raised with any product raised under animal agricultural management conditions, a high profile product safety problem with any farm-raised species has industry-wide implications. Other industries have suffered the consequences of consumer backlash from residue problems, negative media attention and public perceptions of safety problems. These problems can be effectively prevented and avoided through industry-wide proactive quality assurance, total quality management and HACCP based programs for producers and processors. Even imported comparable products tainted with violative residues or contaminants can trigger public attention and suspicion on "all" comparable products regardless of the country of origin. FDA is continuing to develop analytical method tools to detect residues of more drugs and thus can be expected to take an increasingly stronger regulatory role in enforcing proper drug and pesticide use in aquaculture. Again, consumer confidence and trust of an industry's responses and actions to assure the wholesomeness of products will create market advantages both domestically and internationally.

Even though extra-label drug use has been legalized by Congressional legislation, considerable restrictions exist which need to be understood by producers and processors. The veterinarian who prescribes drugs in an extra-label manner is responsible for assuring that no violative residues exist in the marketed product. If scientific information on the human food safety aspect of the use of the drug in food-producing animals is not available, then the veterinarian must take appropriate measures to assure that the animal and its food products will not enter the human food supply. An example may be a drug approved for humans or non-food animals (dogs) may be prescribed extra-label for broodfish because of their intrinsic reproductive value and the fact that they will not enter the human food market. If the same drug is prescribed for food fish then they cannot be marketed for food if human food safety data are lacking to determine a safe withdrawal time.

As the mandatory seafood inspection regulation approaches its time of implementation and industry compliance, the aquaculture sector will be impacted by this new rulemaking. Many large processors have experience with HACCP through their voluntary participation with NMFS's training and product certification programs. However, HACCP is new to most small and medium sized operations that process seafood products, including farm-raised species. HACCP and its potential pre-harvest implications are generally quite unfamiliar topics to most aquaculture producers. There is still a lack of dialogue among the producer, processor and regulatory sectors to evaluate acceptable options to comply with mandatory regulations with flexibility and understanding. The interpretation of existing FDA documents and rulemaking by field inspectors may be different from headquarters staff, specifically the use of the Guide in developing and verifying HACCP plans.

There have been numerous public-private sector partnerships which have proven effective in addressing complex aquaculture issues, including seafood HACCP training, development of QAPs and educational products and progress on new animal drug approvals (80). There is a need for a similar public-private forum and collaboration to address numerous issues specific to the aquaculture community involving the pending seafood HACCP regulations and related matters to avoid undue confusion and burdens.

RECOMMENDATIONS

There are numerous pending issues that require immediate attention, clarification or action by producers, processors and/or regulators to transition smoothly and effectively into the new HACCP regulatory environment for seafood processors. Recommendations may be best addressed by case-by-case options, industry trade organizations, commodity associations, industry-wide actions, media communications and/or regulators. Recommendations are as follows:

- Communication and liaison among aquaculture producers and processors, and FDA regulators should intensify to clarify uncertainties and acceptable protocols regarding reasonable options for processors to address pre-harvest, on-farm safety concerns in their HACCP plans. Industry should submit concerns in writing to FDA and request

clarification uncertain issues that may have varying interpretations or applications. The official FDA responses should be broadly disseminated to further educate the industry and FDA/state regulatory field staff.

- The aquaculture industry should take a proactive approach before HACCP regulations go into effect to discuss exactly what information producers may need to provide, how to make it available and what monitoring and verification testing should be conducted. These issues need to be addressed now by producers and processors to arrive at adequate starting points that are reasonable to all parties for assuring product safety in processor HACCP plans. This may begin with conferences or workshops at the state, regional or national levels. FDA's guidance should be solicited as needed. Processors may do more testing if adequate safety assurance controls and records are not provided by producers.
- A key issue is whether a raw, incoming product from a farm is the first CCP in a processor's HACCP plan and/or that producers are expected to employ standard operating procedures (SOPs) on the farm as preventative measures to avoid product contamination as suggested in a QAP, should be evaluated by the industry because the outcome may vary on a case by case basis depending on the processor, the reputation of the producer and the recognition of the producer QAP.
- Processors need to fully understand the production side of aquaculture and employ sound knowledge to decide what CCPs are reasonably associated with pre-harvest conditions on a case-by-case basis. This is important to minimize resource expenditures yet assure product safety and compliance with the HACCP regulations. The ultimate party responsible for assuring product safety and consumer confidence is the processor yet all segments of the food industry share in this responsibility.
- Producers likewise need to understand the responsibility and role that processors have under the new seafood inspection regulations and realize their responsibility with processors in assuring the pre-harvest safety of their incoming raw products in the farm to fork food spectrum.
- Industry commodity associations and national organizations should begin developing guidance and offering assistance to producers and processors to encourage dialogue which addresses compliance with the new HACCP regulations. Reasonable options for recordkeeping systems, verification testing, acceptance and use of QAPs, use of certificates of producer compliance and other related information should be developed and broadly disseminated.
- The aquaculture industry and community should become familiar with the FDA publication, "Fish & Fisheries Products Hazards & Controls Guide", and provide written comments for the next revision of this document that address the value of QAPs, verification testing and reasonable options to avoid contaminants and assure product safety.

- Recordkeeping systems that record, store and report critical on-farm safety-related information by production units and fish lots harvested should be developed as user-friendly software packages and hardcopy formats for use by aquaculture producers. These systems and formats should be deemed acceptable by FDA and processor organizations to meet either SOP or CCP documentation. Industry can develop reporting standards for producer adoption to assist them with meeting any HACCP-related pre-harvesting reporting requirements.
- The need for verification testing should be addressed by processors and producers based on residue risk factors, FDA guidance and practical approaches. This should also include the need for third-party verification that may be sought by some processors. Third party verification can be used if processors need assurances that producers are doing what they say they are doing. Who can or should be a competent third party verifier if needed? Current residue monitoring studies have revealed no chemical residue safety risks, although preventative practices are always needed.
- An info brief should be developed jointly by the aquaculture industry and FDA which provides some guidelines, points of clarification and expectations regarding options for producer actions and processor compliance with farm-raised products. This information should be broadly disseminated among the industry and regulatory communities.
- Producer and commodity associations at national and state levels should include HACCP topics and issues as presentations at annual meetings to facilitate education of both processors and producers. A HACCP panel composed of local regulators, extension specialists and industry representatives from the processing and production sectors would be an effective educational format.
- The aquaculture industry should recognize and support the concept that the best approach to reduce potential foodborne illnesses and safety risks to consumers is an industry-wide farm to fork HACCP-based system. Other animal agriculture sectors which compete for consumer purchases of high protein animal products are proactively supporting such initiatives and will use farm to fork QAPs based on HACCP as a marketing tool. Numerous animal agriculture producer QAPs are being revised to reflect a linkage with HACCP principles.
- FDA surveillance and residue monitoring of imported aquaculture products should continue to create and maintain a level playing field in the marketplace and enforce equivalent standards for safety for all aquaculture products in our nation's seafood supply.
- FDA needs to specify what types of processing operations are covered in the seafood HACCP regulations and what exemptions might exist for small custom processors

at paylake or small farm operations. The seafood HACCP regulations state that there will be no exemption for small processors. The issue lies in the definition of a processor and whether some custom processing operations may be considered retail establishments and therefore exempt from the regulations.

- All producers large and small need to become more educated about QAPs and the mandatory HACCP regulations so all can compete successfully in marketing their products in an increasingly HACCP-based food system. Industry organizations, Extension Services and the aquaculture media should assist with this need.
- Reference to the value and relevancy of producer QAPs should be clearly recognized in FDA HACCP guidance documents, HACCP training materials and media materials directed to producers, processors and consumers.
- Processors should encourage producers to implement industry-sponsored or farm-sponsored QAPs.
- The value and use of commercial rapid field test kits for drug or pesticide residues in fish need to be validated as effective and credible tools for verification testing. These kits are referenced by FDA in the Guide, yet FDA has not evaluated the efficacy of any products now being marketed. The reliability, cost and rapidness of field screening tests should be evaluated against quantitative laboratory tests.
- Academia should become knowledgeable of the pending HACCP-related changes and incorporate the implications of these changes in teaching and research. Broader expertise will be needed in food safety within the Cooperative Extension Service and Sea Grant Marine Advisory Service to work with both producers and processors and train others to provide industry services as needed.
- Public and private sectors should cost-share and support the revision of the publication, "Guide to Drug, Vaccine and Pesticide Use in Aquaculture." This publication was first published in 1994 and needs updating, especially the section on pesticides. This publication has been termed the "bible" by Food Chemical News because the content has been reviewed and approved by the three regulatory agencies EPA, FDA and APHIS. This publication is a valuable addition to a QAP and should be used and referenced by anyone involved in U.S. aquaculture.
- Acceptable production practices regarding the use of therapeutants, pesticides and other products to control aquatic weeds, combat disease outbreaks or other management actions with safety implications at the farm level should be reviewed by processors and regulators to be adequate for HACCP requirements through producer QAPs or comparable producer actions.

- There is a short time from the farm to the processing plant with live products. Also unpredictable off-flavor problems can affect when fish may be acceptable for processing. Because of the short time involved with live fish moving from the farm to the processor any HACCP plan needs to be sensitive to the importance of timeliness and avoid undue delays in this farm to dockside transfer.
- Producers involved with growing row crops or other non-aquatic crops need to avoid any cross-contamination of aquaculture production units from insecticide or herbicide drift. Aerial applicators should be especially warned of risks associated with spraying in the vicinity of aquaculture operations and adhere to ample buffer zones. Trace pesticide residues found to date are associated with non-aquacultural production practices.
- The concern about residues of persistent organochloride pesticides in soils or environments associated with aquaculture production sites in areas previously used to grow row crops historically treated with such compounds should be addressed. These safety risks are extremely low and no cases of violative residues in farm-raised products from this source are known to have been reported, yet regulators continue to consider this an existing risk. Education is needed backed with evidence of safety from existing data and avoidance of potential hot spot locations.
- Laboratories need to be identified that use acceptable GLPs and testing protocols to sample and test various aquaculture species for residues of different compounds of highest regulatory concern if warranted.

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PRELIMINARY DESIGN OF A SEAFOOD HACCP MONITORING SYSTEM

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INTRODUCTION

The United States Food and Drug Administration (FDA) has announced their intent to establish a new mandatory system for the inspection of seafood based upon the Hazard Analysis Critical Control Point (HACCP) system (FDA, 1994). The program, which goes into effect on December 18, 1997, will require all seafood processors, packers and importers to develop and maintain a preventative system to ensure the safety of seafood consumed in the United States. According to FDA, each HACCP plan must identify the potential safety hazards, determine the critical points in processing and handling where these hazards can be controlled, identify critical limits for each control point, outline corrective actions, and establish process monitoring and verification procedures.

In an effort to smooth the transition to this new regulatory environment, FDA has specified the appropriate course of action for each step in seafood processing operations, from a regulatory viewpoint (Ward *et. al*, 1996). However, FDA has suggested multiple methods for monitoring, record keeping, and verification of HACCP plans. Although the process monitoring techniques currently in practice in the seafood industry are adequate, the application of automated process monitoring and control technologies provides the potential to greatly simplify many tedious and time consuming tasks required for HACCP. A computer is ideally suited for performing multiple, repetitive tasks. It can be programmed to continuously monitor critical control points for each day of operation. Furthermore, a computer can be used to generate and store reports using acquired data.

The overall goal of this research was to develop and demonstrate an automated process monitoring and record keeping system for the seafood industry to be used in complying with mandatory HACCP based regulations. However, the wide diversity of seafood products necessitated the selection of a single commodity for system development. Therefore, specific objectives included:

1. The design of a critical control point monitoring system which allowed blue crab (*Callinectes sapidus*) processing unit operation temperatures to be recorded and maintained without disrupting the established process;
2. The development of an automatic HACCP report generator capable of providing the summary information required to verify regulatory compliance and allow seafood processors to evaluate the operation of their facilities.

METHODS AND MATERIALS

The seafood HACCP monitoring system consisted of two primary sub-systems which worked in conjunction with each other: a data acquisition system capable of monitoring the process critical control points, and a data analysis/reporting system which analyzed control point data obtained for each unit operation and generated process status and FDA compliance reports. The data acquisition system (Figure 1) tracked individual 2000 pound batches of blue crab as they were processed.

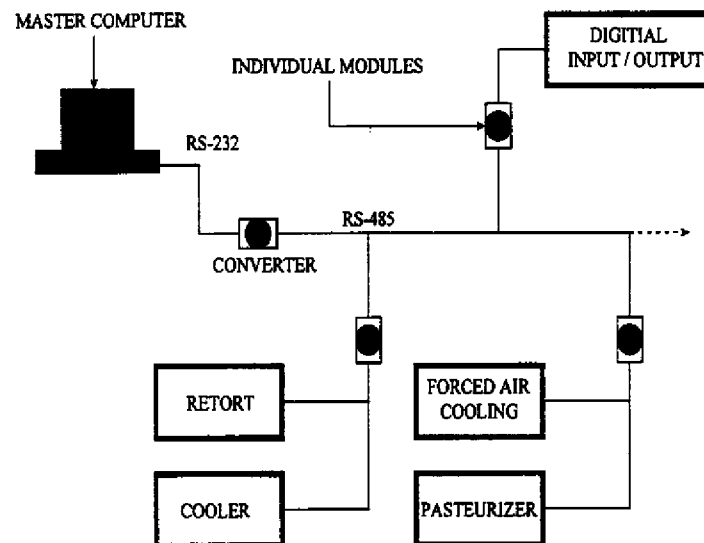


Figure .

When the raw product arrived at the processing facility, the crabs were sized and then placed in a large stainless steel cart until 2000 pounds were in each batch. Each had wheels to allow the batch to be moved from operation to operation. Full carts were pushed inside the retort for cooking. After cooking, the cart was allowed to cool at ambient temperature for not more than two hours and finally was placed into a refrigerated cooler, where the product awaited picking. Following overnight cooling, the cart was pushed into the picking room where the crabs were backed and the meat was picked from the shell. The meat was sorted into different grades and then each container was carefully weighed and packed on ice until it was shipped to market. If the meat was to be pasteurized, it was bagged or canned,

pasteurized, and then shipped to market.

A network interface was installed at each process operation to accommodate the moving batches. The interfaces were designed as a distributed data acquisition system employing a Remote Sensor-to-Computer Interface (RSCI), and consisted of a master computer connected to multiple remote micro-controllers. The data acquisition modules communicated with the master computer through an RS-232 to RS-485 converter and the RS-232 serial port. The remote sensors were connected in a daisy-chain configuration and RS-485 protocol was used to reduce signal distortion and accommodate any future expansion needs, with minimal additional wiring. The RSCI system allowed sensors to be located nearest the event of interest, further reducing signal distortion and improving signal quality.

Temperatures were monitored at four unit operation locations: (1) retorting (2) forced ambient air cooling (3) pre-pick cooling and (4) pasteurization. Pre-pick cooler doors were also instrumented to monitor the time of day which they were opened and closed and determine the effect on cooler room temperature. Each of these unit operations were selected for monitoring because of their importance in hazard prevention.

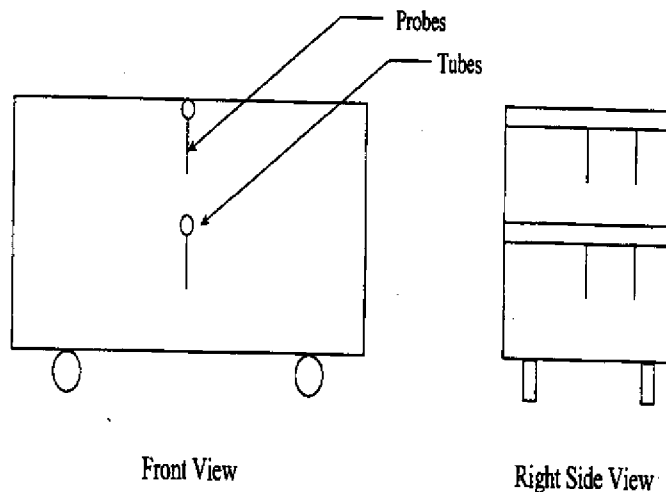
The data analysis/reporting system consisted of software written specifically for HACCP monitoring and reporting. Temperature data recorded at each unit operation were analyzed to prepare a summary report including maximum, minimum, and mean temperature. Daily operational summaries could be printed and signed by HACCP certified personnel and the printed copy stored as verification of HACCP plan operation. The software also permitted electronic storage of process data, allowing plant operators to display of acquired data in a spreadsheet format as well as create time-temperature graphs to assist in evaluating thermal process operations.

System Hardware

The monitoring system (refer to Figure 1 above) was controlled by a personal computer utilizing a 80486 DX based processor with a 33 MHz clock and 8 MB of RAM. A standard serial cable interfaced the computer to a RS-232 to RS-485 converter module (American Advantech Corp., 750 East Arques Ave., Sunnyvale, CA 94086). RSCI modules were powered by a 24 DC volt power supply (American Advantech, Part Number PWR-245), which was centrally located with the computer and the RS-232 to RS-485 converter. A communication cable, containing four shielded conductors (Allied Electronics, Manufacturer's Type 8434), connected the converter module to other system data acquisition modules located at each process operation. System sensor data were acquired with American Advantech ADAM series data acquisition modules (American Advantech, Part Numbers ADAM-4011, ADAM-4018, ADAM-4050).

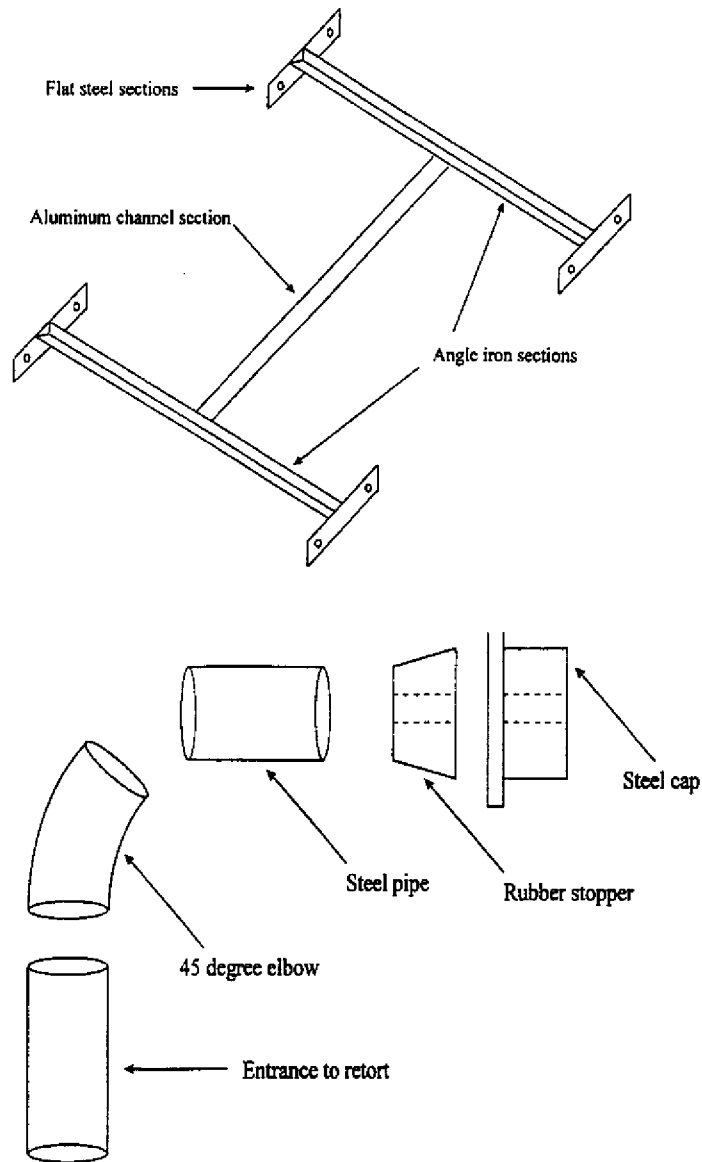
Type T thermocouples (Omega Engineering, Catalog Number TT-T-24) were used to measure temperatures at each unit operation. Type T transition junction probes with grounded junctions and stainless steel sheaths (Omega Engineering, Catalog Number TMTSS-125(G)-6) were soldered to thermocouple wires for recording the product

temperatures. Individual thermocouple probes were fitted with male OST connectors (Omega Engineering, Catalog Number OST-T-MF) and permanently mounted on the carts used to transport blue crabs between each operation (Figure 2).

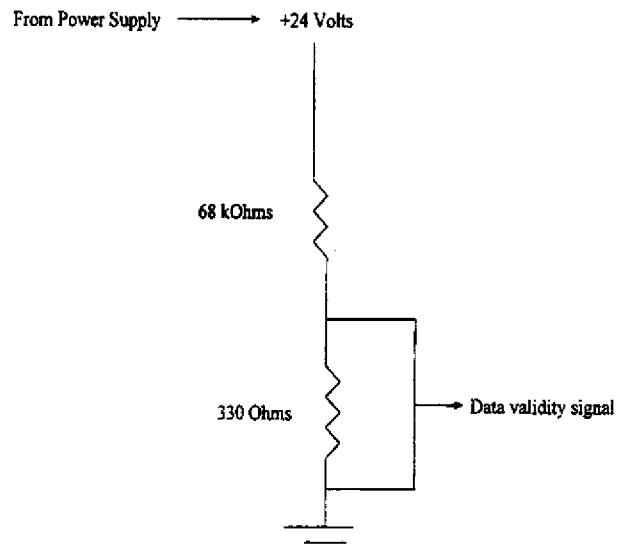
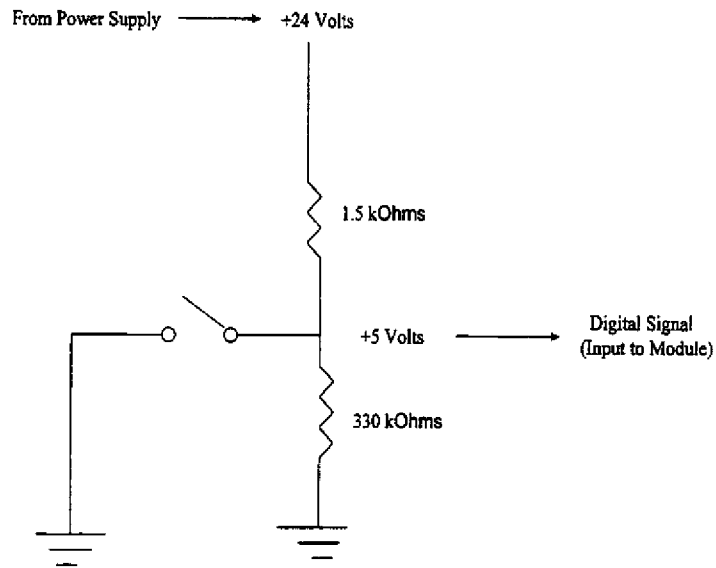


Stainless steel tubing was used to protect thermocouple probes and ensure their proper positioning inside each cart. Two tubes were placed horizontally across the width of each processing cart. A pair of holes drilled in the wall of each tube allowed thermocouple wires to be placed inside. Each tube contained two thermocouples with one end fitted with a male OST connector and the other end soldered to a thermocouple probe. Tube sections were threaded on each end and secured to the cart frame with stainless steel washers and nuts. Thermocouples were connected to the data acquisition system at monitoring stations located at the forced ambient air cooling, pre-pick cooling and pasteurization operations.

Inside the retort, thermocouples were connected to the system through OST plugs mounted in a self-supporting frame (Figure 3). The frame was made of two sections of angle iron spanning the width of the retort. An aluminum channel concealing thermocouple wires spanned the length of the frame. The angle iron was held in place by flat steel welded to each end. Flat steel sections contained two holes onto which stainless steel nuts were welded, permitting pointed stainless steel bolts to anchor the frame inside as a compression device. Thermocouple signal lines entered the retort through a custom constructed pipe fitting (Figure 4) which was comprised of a steel cap, a steel pipe, a 45° elbow, and a rubber stopper.



Digital signals acquired by the system included the status of pre-pick cooler doors and a data validity signal used to indicate that an instrumented blue crab cart was connected to a system unit operation. Pre-pick cooler doors were monitored with digital signals generated through switches mounted inside the door frames. Each switch was connected with the signal conditioning circuit (Figure 5) which connected to an ADAM series digital input module. The data validity indicator consisted of a thermocouple connected to a circuit (Figure 6) producing a signal voltage change independent of temperature which could be sampled through a thermocouple channel. Changes in the data validity voltage were translated with software to digital on/off signals.

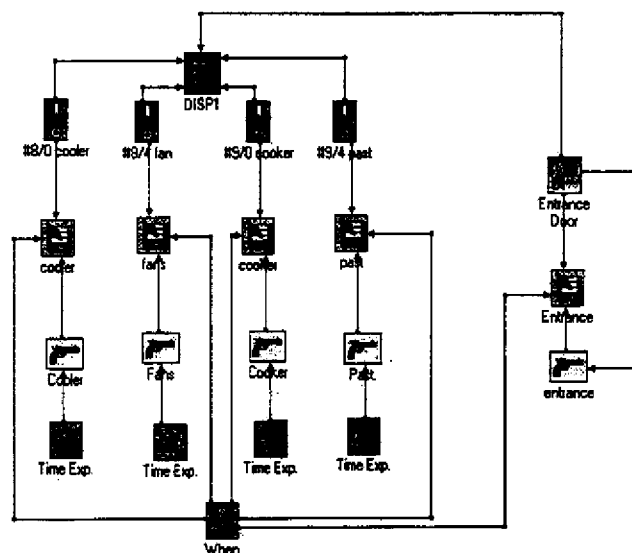


System Software

The software packages used to operate the seafood HACCP monitor consisted of a data acquisition package (GENIE, American Advantech) to poll each RSCI module for data collection and storage, and the previously discussed data analysis/reporting system software.

GENIE is an object-oriented data acquisition and control package equipped with a graphical user interface (GUI) that allows the user to create control strategies through a library of standard mathematical and control functions. The user selects an icon and drops it on the work space with several manipulations of the mouse. Data flow was controlled by connecting icons, allowing the user to visualize the data stream. Using a display designer function, users also create simple numerical displays that allowed real-time display of the temperatures being monitored.

A customized data acquisition program was designed as shown in Figure 7. The thermometer icons represent individual thermocouple probes. Each monitored location contained four individual probes, two monitoring product temperatures, one monitoring ambient temperature, and one serving as a data validity signal. Although the HACCP monitoring system operates continuously, data only needs to be stored when a unit operation contains product. Therefore, the system was designed to collect data only when material is being processed. This was accomplished through the data validity signal. When an instrumented cart was connected at a unit operation of interest, the data validity signal stored a one (1) in the data file. If no batch was present, a zero (0) was stored.



The data acquisition program was capable of sampling temperature data at 1 Hertz (Hz). However, the rate of temperature change at each unit operation varied. Using GENIE, a data file was created for each unit operation. The ASCII files recorded the date, time, data validity signal, and temperature data for each unit operation at specified intervals. In the pre-pick cooler, the computer monitored temperatures at 1 Hz for 15 minute intervals and stored an average value. In the forced air cooling operation and the pasteurization operations, actual temperatures were stored at one minute intervals. In the cooker, actual temperatures were stored at 20 second intervals.

GENIE monitored the pre-pick cooler door status signals at a rate of 1 Hz. However, to prevent large volumes of data from accumulating, each reading was not stored. When a door was opened, the time, date and status of the door were stored once each second. When a door was closed, data collection ceased. By examining the times stored in the log file, the duration of each door opening event could be determined along with its influence on the temperature of products stored in the cooler.

Experimental Procedures

Performance of the seafood HACCP monitor was evaluated by installing the system in a commercial processing environment and conducting two tests designed to demonstrate its operation. The first confirmed the accuracy of the information acquired. Thermocouple probes were plugged in at each unit operation and placed in an ice bath followed by a boiling water bath while the system was acquiring data. Control ice and boiling water bath temperatures were simultaneously measured with an electronic thermometer. In the second experiment, the operation of the HACCP monitoring system during blue crab processing was tested by allowing it to run as designed and analyzing the data and reports obtained from the retort and pre-pick cooler operations for a representative 2000 lb batch.

RESULTS AND DISCUSSION

Testing of the seafood HACCP monitor revealed important information involving both the system's operational characteristics and the function of process operations being monitored. In general, the monitoring system met the overall project objectives of allowing process temperatures to be non-intrusively monitored and recorded automatically and allowing this information to be summarized for verification of HACCP operation. The system operated continuously, displaying process temperatures to plant operators in real time while recording the information for subsequent analysis. In addition to real time display, the system was capable of analyzing stored data and generating operational summaries while simultaneously conducting monitoring functions. The analysis software was capable of displaying recorded data in spreadsheet format, generating a time-temperature history, or a unit operation summary.

During the operational test, it was noted that care was needed in the handling of individual OST thermocouple connectors at each monitoring station. As designed, the wire/connector junction was not strong enough to ensure that the wire did not separate from the connector during normal operation. A loop of thermocouple wire was subsequently incorporated into each connector along with a metal restraint which acted as a strain relief mechanism to minimize this problem and ensure data integrity.

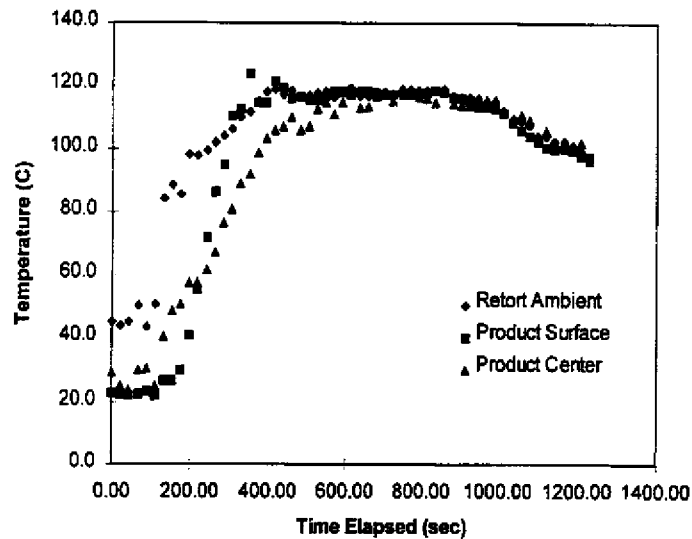
Accuracy Testing

Data collected during the seafood HACCP monitoring system's accuracy tests were

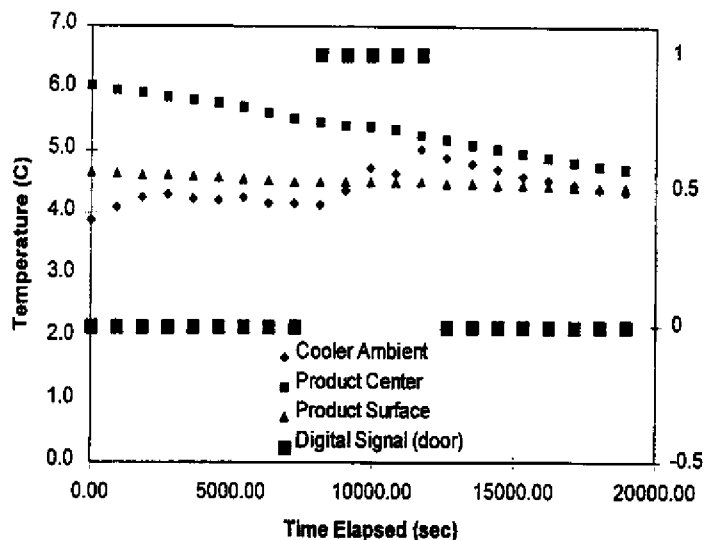
compared and statistically analyzed to determine if the temperatures measured by the system were precise. Sensed temperature readings were within 1 °C of control readings. Correlation coefficients of 0.964 or higher were calculated for all comparisons.

Operational Testing

Temperature data was obtained during retort processing of a 2000 lb batch of blue crab and analyzed with the report generator (Figure 8). It is clear from these measurements that the cook administered to the product was not uniform, with the temperature at the surface reaching a maximum at more than twice the rate of those in the center. The summary report showed the ambient temperature inside the retort was 43.5 °C prior to cooking, and reached a level of 119.0 °C 330 seconds following the introduction of steam into the retort. Temperature measurements between individual crabs at the cart s surface showed an initial temperature of 22.1° C which rose to 124.0 °C 240 seconds following steam introduction. In the cart center, an initial temperature of 23.8° C was recorded which climbed to 118.0 °C 636 seconds into the cooking process.



Data recorded by the system for 2000 lbs. of blue crabs stored in the pre-pick cooler are shown in Figure 9. Note that the ambient temperature measurements rise between the times of 8000 and 12000 seconds. During this time period, the cooler doors were opened, allowing warm air to enter the room. However, product temperatures were not increased by the rise in cooler ambient temperature, and continued to approach or remain below the 4.8 °C target level.



SUMMARY AND CONCLUSIONS

A personal computer based temperature monitoring system has been developed which allowed unit operations within a seafood processing facility to be continuously monitored. The system allowed product and ambient temperatures to be monitored without physically disturbing processing equipment or affecting established processes. The temperature data were acquired at various sampling rates, based upon the length of the process and rate of temperature change. Acquired data was stored for subsequent analysis and summarization needed for FDA seafood HACCP based regulation compliance. System testing revealed the wide range of temperatures which might be encountered in a typical 2000 lb batch of blue crab and demonstrated that it is important to monitor a variety of temperatures in the product if HACCP plan operation is to be accurately verified. Future development efforts concerning this system will be directed at improving the individual sensor connections to the system and modifying the analysis software to include a check sheet for entering other HACCP operational data for electronic storage and reporting.

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THE QUALITY ASSESSMENT OF SEAFOOD PRODUCTS SOLD IN FLORIDA BY ORGANOLEPTIC, CHEMICAL, AND MICROBIOLOGICAL METHODS

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INTRODUCTION

During the past decade, the U.S. population has become more health-conscious and as a result is paying closer attention to human nutrition. As a direct consequence, consumption of seafood, a relative low fat food, is up twenty percent over the past ten years and the trend is continuing to grow. The increasing demand for seafood is due largely to the public's acceptance of the health benefits of including seafood in the diet. Studies have proven that seafood is not only a lean, low-fat, and low cholesterol source of protein but also a good source of vitamins and minerals.

The recent reports on the outbreak of seafood-associated food borne illness has raised general public's concern about the safety of these products. According to the statistics of Center for Disease Control (CDC), fish and shellfish were responsible for 10.5% of all outbreaks of foodborne disease reported for 1978-1987 (CDC, 1989). Consumer Reports (1992) also states that seafood safety and quality has been emerging as a paramount concern for commercial, regulatory and public interests in the United States. The Food Safety program at the State of Florida is designed to protect the public health from injury by product use or from injury resulting from misrepresentation. Therefore, the objective of this study was to evaluate and assess the quality and safety profiles of retail seafood products (shrimp and scallop) sold in this state.

MATERIALS AND METHODS

Sample Collection and Delivery

Shrimp and scallop sample used in this study were collected at retail store by the food inspector of the Florida Department of Agriculture and Consumer Services. Sample was

either kept in frozen state or packed with BLUE ICE (Rubbermaid Specialty Products Inc.; Wooster, OH) and shipped directly from the collection site to Food Laboratory (Tallahassee, FL) for testing. Upon the arrival at laboratory, sample was immediately stored in freezer (-20 °C) or refrigerator (4 °C) until analysis. Sample was prioritized to microbiological assays preceding to the organoleptic evaluation and chemical analyses. All testing was initiated within 24 hr following the reception of sample by laboratory.

Determination of Shrimp (Scallop) Material in Breaded Shrimp (Scallop)

Determination of meat material in breaded shrimp was carried out according to the Food and Drug Administration (FDA) regulations §161.175 in Title 21 of the Code of Federal Regulations (CFR, 1996) and FDA Compliance Policy Guides 7108.12 (FDA, 1996). Following weight measurement, shrimp sample to be debreaded was transferred to a container filled with three-fourths of water at 70-80 °F. The wood paddle was suspended inside the container, leaving a clearance of at least 5 inches below the paddle vanes, and spun at 120 rpm for 10 min. The container content was poured onto a pair of stacked-sieves, the ½-inch mesh over the No. 20. The sieves was set under a faucet, preferably with spray attached, and rinsed shrimp with no rubbing of flesh. Shrimp was laid out singly on the sieve as rinsed. Each shrimp was carefully inspected and the remaining breading material was removed by the use of rubber-tipped rod and the spray. After the removal of top sieve, shrimp was drained on a slope for 2 min then transferred to a weighing pan. The contents of the No. 20 sieve was rinsed onto a flat pan and any particles other than breading was collected and added back to shrimp and weighed. The percentage of shrimp material in breaded product was calculated and determined as: $[(\text{weight of debreaded shrimp} / \text{weight of shrimp before debreading}) \times 100] + 2$.

Determination of scallop meat in breaded scallop was performed according to the National Marine Fisheries Service regulations §266.171 in Title 50 of the Code of Federal Regulations (CFR, 1993). Following weight measurement, breaded scallop was individually placed into a water bath maintained at 63 to 86 °F. The breaded sample was allowed to remain at water bath until the breading became soft and could be easily separated from the meat tissue. After this soak-softening process, sample was lightly dry-blotted with a double-layer of thick paper toweling and bread coatings was scraped off from the meat tissue using a spatula or nutpicker. The percentage of scallop meat in breaded sample was calculated and determined as: $(\text{weight of scallop meat} / \text{weight of breaded scallop before debreading}) \times 100$.

Organoleptic Evaluation

Sensory attributes of shrimp and scallop products were examined and evaluated by the qualified organoleptic analysts of Food Laboratory. The evaluation procedure was performed according to the National Marine Fisheries Service regulations §265.104, 265.171, 266.111, 266.121, 266.162, 266.163, 266.164, and 266.165 in Title 50 of the Code of Federal Regulations (CFR, 1993) and FDA Compliance Policy Guides 7108.11 (FDA, 1989). Indole and off-odor (rancidity) were primarily used as the decomposition indicator for the assessment of shrimp and scallop quality, respectively. All organoleptic evaluation was

conducted at room temperature after sample was completely thawed and equilibrated to the room temperature. During evaluation, shrimp was sorted into three classes on the basis of quality status [class I (acceptable), class II (decomposed), or class III (advanced decomposed)]. The percentage of each class was calculated and the quality grade for sample by organoleptic evaluation was then assigned. Shrimp sample was further analyzed by various chemical analyses [pH, ammonium, total volatile basic nitrogen (TVBN), and indole] and these findings were correlated to their sensory characteristics.

pH Determination

Ten grams of peeled shrimp sample was added to a beaker containing 50 mL of distilled, deionized water. The mixture was fully homogenized using a Polytron (PT 3000, Brinkmann) and the pH of suspension solution was measured by a pH meter (Model 410A, Orion).

Moisture Determination

Three to five grams of fresh scallop sample was weighed in an aluminum dish (55 x 15 mm, Fisher Scientific) and dried overnight at 102-105 °C. The weight loss was determined as the moisture content of the sample.

Ammonium Determination

Ammonia chloride solutions (Fisher Scientific) prepared at 10^{-5} to 10^{-1} M were used as the standard to calibrate the pH/Ion meter (Accumet 925, Fisher Scientific). For this experiment, an ammonium ion selective electrode (Fisher Scientific) was chosen and connected to the pH/Ion meter. Five grams of shrimp meat tissue was chopped into small pieces and homogenized with an appropriate amount of water (~ 60 mL) using a Polytron. The shrimp slurry was transferred into a 100 mL volumetric flask and diluted to the volume with distilled, deionized water. One milliliter of ionic strength adjuster (ISA) (Fisher Scientific) was added to the sample solution and the mixture was well-shaken prior to the measurement of conductivity. Corresponding concentration of ammonium in shrimp tissue was determined by extrapolating the conductivity of sample solution from the established calibration curve of ammonium standards of known concentrations.

Determination of Total Volatile Basic Nitrogen (TVBN)

The determination of TVBN in shrimp was performed according to the method of Malle and Poumeyrol (1989). Two-hundred mL of 7.5% (w/v) trichloroacetic acid solution was added to 100 g shrimp meat and the mixture was blended to a homogeneous state. The suspension solution was centrifuged at 2,500 rpm for 10 min. The supernatant collected was filtered through a Whatman No. 1 filter. Distillation of filtrate was performed using the Kjeldahl-type distillator (Pyrex brand, Fisher Scientific) housed on an electromantle (EM 0500/C MK4, Electrothermal Inc.; Gillette, NJ). Briefly, 25 mL of filtrate was loaded into a 500 mL distillation flask containing 6 mL of 10% (w/v) NaOH and distillation was

proceeded till the final volume of 50 mL was obtained in a 125 mL Erlenmeyer flask containing 10 mL of 4% (w/v) boric acid and 0.04 mL of 0.05% (w/v) methyl red and bromocresol green indicator. The collected ammonia (green color) was then titrated with 0.1N sulfuric acid until the neutralization point (pink color) was reached. The TVBN (mg%) was determined as $(Y \times 16.8 \text{ mg of nitrogen})$ per 100 g of shrimp meat tissue, where Y was the volume of 0.1N sulfuric acid consumed by the titration.

Determination of Indole in Shrimp

The determination of indole in shrimp was measured according to the AOAC 981.07 (1990). Shrimp sample subjected to organoleptic evaluation process were sorted into 3 different classes (I, II, or III), respectively. The segregated, peeled (shell-off) shrimp meat was added to the high performance liquid chromatograph (HPLC) grade methanol at a ratio of 1 to 5 (w/v) in a Waring blender. Internal standard, 2-methylindole (Sigma Chemical; St. Louis, MO), prepared in methanol was concurrently added to the blender at the final concentration of 0.10 $\mu\text{g/mL}$. The combined mixture was blended to a homogeneous phase and the suspension was filtered through a 0.45 μm PVDF filter (Gelman Science; Ann Arbor, MI) before injecting into the HPLC. HPLC system consisting of autosampler (712 WISP, Waters; Milford, MA), pump (M-6000, Waters), analytical column ($\mu\text{Bondpack}$, 300 x 4 mm id.; Waters), and spectrofluorometer (Model 470, Waters) was used for this study. Isocratic methanol-water (60+40) was used as the mobile phase and the flow rate was set up at 1.2 mL/min. The fluorescent detection of indole were set up at 280 nm (excitation) and 330 nm (emission), respectively. Standard indole solutions (0.6-7.2 $\mu\text{g}/100 \text{ mL}$; Sigma Chemical) prepared along with 2-methylindole were used to establish the calibration curve. The injection volume for indole standard and shrimp sample extract were 20 μL .

Determination of Aerobic Plate Count (APC)

The APC of seafood sample was determined using the FDA Bacteriological Analytical Manual (BAM) (1992). Fifty grams of thawed sample was aseptically blended with 450 mL sterile diluent for 2 min. The homogenized sample was further diluted to a serial ratio of 10^{-2} , 10^{-3} , and 10^{-4} using the same sterile diluent. Following the dilution step, 1 mL of each diluted sample was pipetted into the petri dish (100 x 15 mm, Fisher Scientific) and thoroughly mixed with the molten plate count agar (Difco Laboratories; Detroit, MI) poured into the plate. After solidification of the agar, petri dish was invertedly incubated for 48 hr at 35 °C. The number of bacterial colonies grown on the plate was counted and recorded.

Isolation and Identification of *Listeria* Species

The modified FSIS method (USDA, 1989) was adopted for this study. Twenty-five grams sample was added to a stomacher bag with 225 mL of the University of Vermont media (UVM) (Difco Laboratories) and homogenized for 1 min. The stomacher bag was sealed with an additional small air pocket and incubated at 30 °C for 24 hr. One-hundred μL of incubated broth was transferred to a Fraser broth and incubated at 35 °C for 24-48 hr. The modified Oxford medium (MOX) plate was streaked with Fraser broth showing darkened culture, and

incubated at 35 °C for 24–48 hr. The suspected colonies of *Listeria* species were selected from the MOX agar plate and inoculated onto a sheep blood overlay agar plate. Following an overnight incubation at 35 °C, plate was examined under the fluorescent lamp and suspected colonies of *Listeria* species were picked with a sterile needle and stabbed to the Motility Test Medium (Difco) and then to brain heart infusion (BHI) broth (Difco). Both inoculations were incubated overnight at room temperature. The BHI cultures were used for (1) Gram staining test, (2) modified Christie-Atkins-Munch-Peterson (CAMP) test with β -lysin disc, (3) streaking BHI agar slants for catalase, oxidase, and MICRO-ID (Organon Teknika Corp., Durham, NC) test, and (4) streaking tryptose agar slants for serology test. The Gram-positive bacteria showing positive toward β -hemolysis was continuously tested through steps (2) to (4). Strain showing negative reaction for β -hemolysis was examined with (a) modified CAMP, (b) carbohydrate (dextrose, mannitol, rhamnose, and xylose) fermentation, and (3) methyl red (MR) tests. The testing procedure of MICRO-ID was performed according to the manufacturer's specifications. Serological assay was performed by testing heat shocked condensed cell culture against antisera specific to *Listeria* strains (*L. monocytogenes* and *L. innocua*) (Difco).

Isolation and Identification of *Salmonella* Species

The procedure of the FDA BAM (1992) was followed to isolate and identify this organism. Twenty-five grams of sample was mixed with 225 mL sterile Butterfield's phosphate buffer in a blender jar. The content was blended at high speed for 2 min before adding with 25 mL of 10X lactose broth. The mixture was well-mixed and incubated at 35 °C for 24 hr. One milliliter of each incubated mixture was transferred to 10 mL of selenite cystine and tetrathionate broth, respectively. The cultured broths were vortexed and incubated at 35 °C for 24 hr. Following incubation, bismuth sulfite (BS), brilliant green (BG), and xylose lysine decarboxylase (XLD) agar plates were streaked with cultures looped from the above two broths. The plates were incubated at 35 °C for 24 hr and examined for the colonies suspected to be *Salmonella*. The suspected colonies of *Salmonella* were selected and inoculated into the triple sugar iron agar (TSIA), lysine iron agar (LIA), and tryptic soy agar (TSA) slants. The inoculated slants were incubated at 35 °C for 24 hr. Microorganism grown in the TSIA and LIA slants were examined for typical *Salmonella* characteristics (BAM, 1992), while culture from the TSA slant was used for the oxidase test. The oxidase-negative culture from TSA slant was further subjected to the API 20E strip (Analytab Products Inc.; Plainview, NY). The procedure of API 20E strip was performed following the manufacturer's instruction.

RESULTS AND DISCUSSION

Table 1 portrays the market form and preparation type of shrimp and scallop products collected for this study. Shrimp and scallop were marketed at either uncooked or cooked state. Uncooked shrimp products included raw salad shrimp, raw breaded shrimp, raw unbreaded shrimp, raw peeled shrimp, all-purpose shrimp, premium shrimp, and frozen shrimp, whereas cooked shrimp products covered peeled cooked shrimp, cooked salad shrimp,

popcorn shrimp, oven crispy shrimp, and fried shrimp. For scallop, uncooked products included bay scallop, frozen scallop, breaded scallop, fresh scallop, and sea scallop. Cooked scallop products only contained popcorn and fried types.

Table 1. Scope and product forms of commercial shrimp and scallop

Shrimp		Scallop	
Uncooked	Cooked	Uncooked	Cooked
raw salad shrimp, raw breaded shrimp, raw peeled shrimp, all-purpose shrimp, premium shrimp, frozen shrimp	peeled cooked shrimp, cooked salad shrimp, popcorn shrimp, oven crispy shrimp, fried shrimp	bay scallop, frozen scallop, breaded scallop, fresh scallop, sea scallop	popcorn scallop, fried scallop

Shrimp (Scallop) Material Determination in Breaded Product

Result for meat material analysis indicates that 48% of frozen, raw, breaded shrimp contained breading more than 55% of its total weight (data not shown). The regulatory guidance for the standard identity of frozen, raw, breaded shrimp as mandated by the FDA (1996) requires the quantity of meat material present in the breaded shrimp not less than 45% of its total weight. Similar non-compliance trend (46%) was observed for the breaded scallop prepared at both cooked and uncooked forms (data not shown). The federal regulation requires that both frozen raw breaded and frozen fried scallop being composed of a minimum of 50% by weight of scallop meat (CFR, 1993).

Organoleptic Evaluation

The quality assessment of shrimp by organoleptic method is shown in Figure 1. Approximately, 95% of cooked, peeled shrimp (n = 90) and breaded shrimp (cooked and uncooked) (n = 23) were rated acceptable in their quality. For raw, peeled shrimp (n = 15), 77% of sample were organoleptically rated unacceptable, which was later verified by the results of pH, ammonium, and indole determinations. The profile for organoleptic evaluation of scallop is illustrated in Figure 2. As result shows 50% of scallop sample (n = 20) tested had been rancid or off-odor.

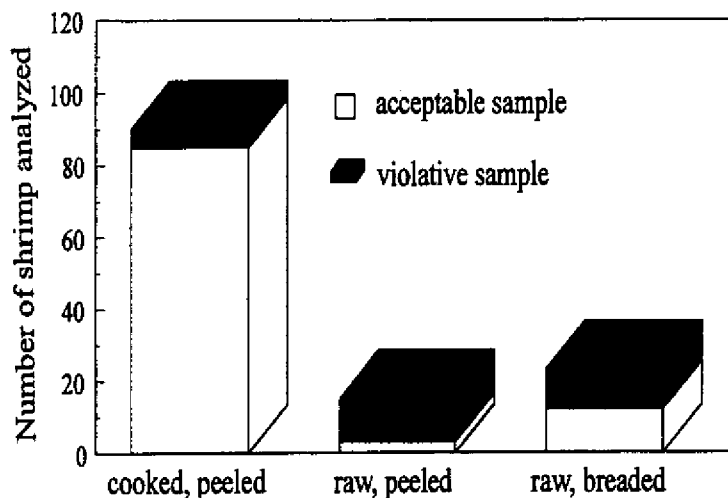


Figure 1. Quality assessment of various shrimp products.

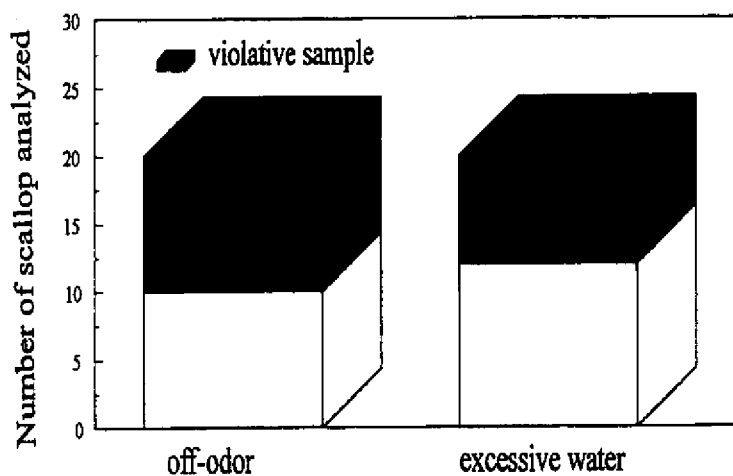


Figure 2. Quality assessment of scallop

pH Measurement

The pH measurement indicated that shrimp with acceptable quality yielded a neutral (7.1-7.2) pH while alkaline value (\geq pH 8) was obtained for the unacceptable ones. The rise in pH was primarily attributed to the breakdown of amino acids by bacterial enzymes to various amine compounds and further deamination. Normally, fresh shrimp with acceptable quality will give a pH value around 7.0 to 7.2. Similar findings were also observed for the scallop products (data not shown).

Moisture Determination

The moisture profile of fresh scallop is shown in Figure 2 above. Forty percent of fresh scallop ($n = 20$) tested was found to contain moisture content higher than 88%. The finding of high amount of water in fresh scallop indirectly implies the extra use of sodium tripolyphosphate (STP) applied to these products. Normally, fresh scallop is composed of approximately 80-84% moisture content. STP has been widely used to retain moisture content of seafood product during refrigeration storage. According the FDA Interim Policy Guide, the scallop product would be deemed adulterated if it contains a moisture content greater than 84% (Anonymous, 1993).

Ammonium Determination

Ammonium has been used as a parameter to reflect the quality status of shrimp. Our study showed that ammonium result had a high correlation to the findings of organoleptic evaluation. All raw, peeled shrimp with unacceptable quality had very high ammonium content. Ammonium ranging from 350 to 650 ppm, with an average of 500 ppm, were found in these decomposed shrimp. Fresh shrimp, contrary to these decomposed products, showed 14 ppm ammonium content in its tissue.

Determination of TVBN

Although the P value [ratio of trimethylamine (TMA)/TVBN] was cited as a good index to determine the freshness (decomposition) status of seafish (herring, cod, whiting, and mackerel) by Malle and Poumeyrol (1989), the relationship of single TVBN value to organoleptic evaluation in decomposed shrimp was not highly correlated as what was expected. Study showed that decomposed shrimp carried TVBN value ranging from 18 to 40 mg nitrogen per 100 g shrimp meat tissue. TVBN values ranging from 20 to 25 mg nitrogen per 100 g meat tissue, however, was found in other frozen shrimp with acceptable quality. Fresh shrimp used as control in this study showed a TVBN value of 30 mg nitrogen/100 g shrimp meat tissue. It seems apparently that P value is more applicable in determining freshness of pelagic fin fish rather than in shellfish.

Determination of Indole in Shrimp

The high performance liquid chromatographic profile of indole standard along with 2-methylindole (internal standard) is shown in Figure 3. Indole and 2-methylindole distinctly eluted at 6.30 and 8.02 min, respectively. Relative correlation coefficient for the calibration standards (0.6 to 7.2 $\mu\text{g}/100\text{ mL}$) was 0.999 (data not shown). When decomposed shrimp extract was injected and analyzed by the HPLC, two well separated peaks migrated at 6.27 and 7.95 min were observed on the chromatogram (Fig. 4). Peak with retention time of 6.27 min was identical to that of standard and identified as indole. Shrimp, which organoleptically rated as decomposed grade, was consistently verified by the HPLC results. Most of class III sample were found to contain indole at the range of 50-75 $\mu\text{g}/100\text{ g}$ meat tissue. Some severely decomposed shrimp sample were found to have indole level higher than 100 $\mu\text{g}/100$

g meat tissue. A moderate level (20-30 $\mu\text{g}/100$ g meat tissue) of indole was found in class II shrimp, while shrimp rated as class I was found to contain indole less than 25 $\mu\text{g}/100$ g meat tissue. No indole was detected in the fresh shrimp used as control.

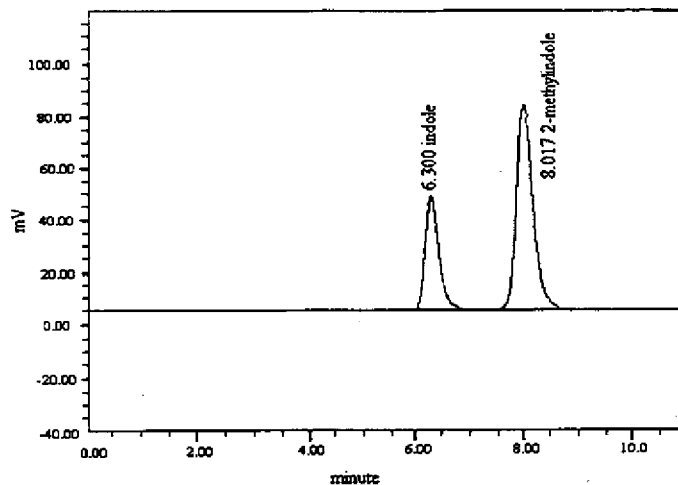


Figure 3. High performance liquid chromatographic profile of indole and 2-methylindole

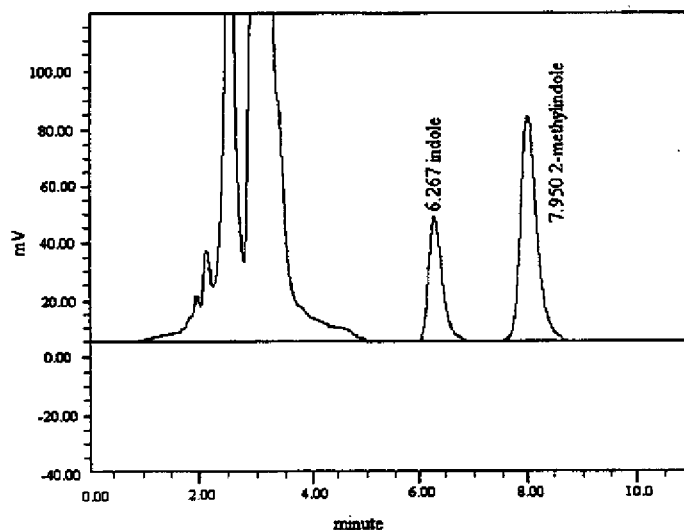


Figure 4. High performance liquid chromatographic profile of shrimp tissue extract.

Determination of APC

The profile of APC number for shrimp and scallop is shown in Figure 5. Study data indicated approximately 62% of sample ($n = 100$) analyzed had APC levels less than 3,000/g. None of sample was found to have APC number higher than 1.5 million.

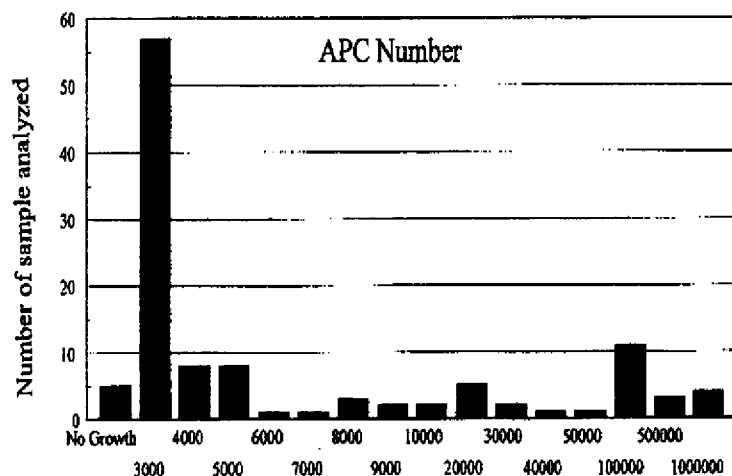


Figure 5. Aerobic plate count (APC) profile of seafood product.

Isolation and Identification of Pathogenic Bacteria

The screening for pathogenic *Salmonella* and *Listeria* species in shrimp and scallop samples indicated the absence of these organisms from these products (data not shown). Although none of *Salmonella* species was found in these products, 10 samples tested positive for *Listeria* species (Table 2). Of 10 contaminated samples, 6 were uncooked shrimp and 2 each were cooked shrimp and scallop, respectively. Three vendors were found to have more than one of their products contaminated with *Listeria* species. Though the contaminating bacteria was later identified and verified as non-pathogenic strain, it did imply the possible product cross-contamination during processing or insanitary manufacturing conditions.

Table 2 Shrimp and scallop products contaminated with *Listeria* species

Product	Vendor	<i>Listeria</i> species
raw breaded shrimp	S1	positive
oven crispy shrimp	B1	positive
raw peeled shrimp	S2	positive
raw peeled shrimp	S2	positive
popcorn scallop	S1	positive
cooked salad shrimp	B2	positive
frozen shrimp	F1	positive
frozen shrimp	N	positive
popcorn scallop	S1	positive
peeled shrimp	N	positive

CONCLUSIONS

Survey result showed nearly half of breaded shrimp and scallop tested in this study contained more than 50% breading. The majority (77%) of raw, peeled shrimp were organoleptically rated as decomposed and unacceptable, which was verified by the pH, ammonium, and indole determinations. In addition to off-odor (rancidity), excessive amount of water found in fresh scallop implies the extra use of phosphate for economic adulteration. Microbiological data showed 50% of the shrimp and scallop samples tested had aerobic plate count (APC) levels less than 3,000/g and were free from contamination of *Salmonella* spp. and pathogenic *Listeria* species.

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COMPOSITIONAL CHARACTERISTICS AND LIPID CLASSES OF GREY SEAL

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INTRODUCTION

Grey seals (*Halichoerus grypus*) are found in the Atlantic waters from Labrador to Nova Scotia, but congregate on islands in the Gulf of St. Lawrence to breed. Currently, grey seals are not commercially harvested, however, they have a negative impact on Canadian commercial fisheries. They damage fishing gear, compete with fisherman for fish, and transmit parasites to cod and flatfish flesh that must be removed during processing (Malouf, 1986). Therefore, there is an interest in controlling the grey seal population either through a commercial harvest or by culling.

The fatty acid composition of the blubber of several species of seal, including grey seal, has been documented (Ackman, 1965;1971; Jangaard, 1968; Kakela, 1993; Shahidi, 1991; 1993; 1994; 1995; West, 1979); however, other tissues of most species of seal have not been examined. Lipid classes of the Mediterranean Monk Seal have been determined, but such studies have not been carried out for other species of seal (Henderson, 1994).

This paper presents data on the omega-3 fatty acid composition of blubber, muscle, heart, liver, kidney, and brain of grey seal. Lipid classes of selected tissues are also provided.

MATERIALS AND METHODS

Samples were obtained from three adult grey seals harvested in Nova Scotia in June of 1995. Tissue samples were homogenized, vacuum packaged and stored at -26°C until used (usually within 30 days).

Moisture content was determined by oven drying of 2-5 g sample at 105°C to a constant weight (AOAC, 1990). Ash and total nitrogen were determined by the AOAC (1990) methods and the crude protein content was calculated as total nitrogen x 6.25. Total lipids were extracted and determined by the procedure of Bligh and Dyer (1959).

For fatty acid analysis, crude lipids, extracted by the method of Bligh and Dyer (1959), were converted to fatty acid methyl esters (FAMES) by transmethylation in acidified methanol (Keough, 1987). FAMES were then separated on a 30m x 0.25mm i.d. fused silica capillary column (Supelco, Oakville, ON) using a Perkin Elmer 8500 gas chromatograph. Oven temperature was set initially 220°C for 10.25 min and then ramped to 240°C at 30°C/min and held there for 9.00 min. The injection port and flame ionization detector were at 250°C.

Lipid classes in different tissues of grey seal were determined using the Iatroscan MK V TLC/FID Analyzer (S.P.E. Limited, Concord, ON). A 1 mL aliquot of sample in chloroform/methanol (2:1, v/v) was spotted on the Chromarod-SII. Development was conducted using hexane/diethyl ether/ formic acid (85:15:1, v/v/v) for separating the nonpolar lipids and then chloroform/methanol/water (80:35:3, v/v/v) was used to separate polar lipids, as described by Innis and Clandinin (1980).

RESULTS AND DISCUSSION

Proximate Composition

Figure 1 represents the proximate composition of selected grey seal tissues. As expected, blubber had the highest total lipid content, followed by brain, liver, heart, kidney, and muscle. Muscle had the highest protein level and brain was highest in its moisture content. Ash content was less than two percent for all tissues examined.

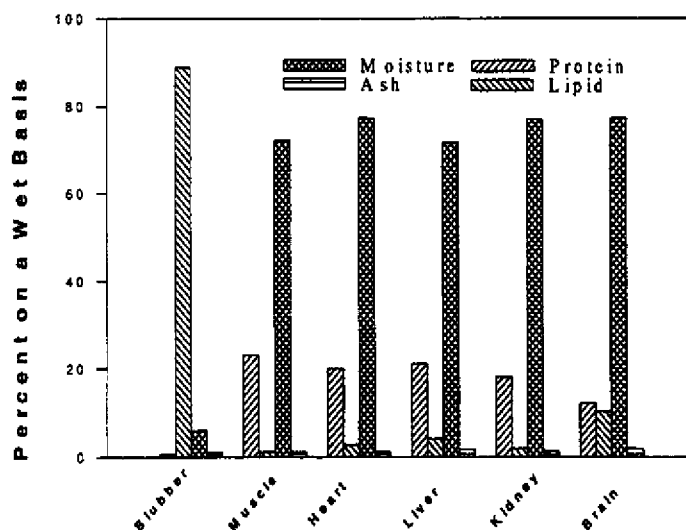


Figure 1. Proximate composition of selected tissues of grey seal.

Fatty Acid Composition

Table 1 provides data on the content of selected fatty acids in the various tissues of grey seal. Oleic acid (18:1 ω 9) was the dominant fatty acid in all tissues, except liver where stearic acid (18:0) was abundant. Arachidonic acid (20:4 ω 6) was present in very low quantities in blubber (0.51%), but was present in much higher amounts in other tissues with heart (11.04%) being the richest source.

Table 1. Content of Selected Fatty Acids of Different Tissues of Grey Seal (% of Total Lipids)

Fatty Acid	Blubber	Muscle	Kidney	Heart	Liver	Brain
18:0	0.94 \pm 0.02	6.20 \pm 0.20	10.44 \pm 0.08	9.07 \pm 0.04	14.94 \pm 0.51	16.33 \pm 0.55
18:1 ω 9	24.50 \pm 0.44	17.48 \pm 0.48	15.91 \pm 0.31	17.46 \pm 0.24	12.79 \pm 0.40	17.25 \pm 0.61
20:4 ω 6	0.51 \pm 0.00	4.31 \pm 0.09	10.78 \pm 0.21	11.04 \pm 0.20	9.28 \pm 0.30	5.51 \pm 0.04
20:5 ω 3	4.85 \pm 0.13	5.60 \pm 0.14	6.46 \pm 0.47	6.92 \pm 0.06	9.24 \pm 0.33	0.34 \pm 0.00
22:5 ω 3	5.06 \pm 0.05	2.94 \pm 0.25	2.26 \pm 0.03	1.62 \pm 0.03	4.11 \pm 0.12	3.25 \pm 0.10
22:6 ω 3	8.91 \pm 0.29	9.30 \pm 0.82	3.35 \pm 1.22	6.08 \pm 0.85	6.69 \pm 0.44	13.31 \pm 0.45
Total ω 3	18.82 \pm 0.47	17.83 \pm 0.96	12.07 \pm 0.75	14.62 \pm 0.76	20.04 \pm 0.01	16.90 \pm 0.54
Total ω 6	1.84 \pm 0.09	6.39 \pm 0.15	14.31 \pm 0.84	15.06 \pm 0.31	12.89 \pm 0.53	7.76 \pm 0.02
ω 6/ ω 3	0.10 \pm 0.01	0.36 \pm 0.02	1.19 \pm 0.06	1.03 \pm 0.03	0.64 \pm 0.03	0.46 \pm 0.01

The content of saturated, monounsaturated, and polyunsaturated fatty acids (PUFA) in different tissues is provided in Figure 2. Saturated fatty acids were highest in the brain (36.76%), and lowest in the blubber (14.82%). The monounsaturated fatty acids were lowest in the liver (24.80%), but were present in higher amounts in the brain, heart, kidney, muscle, and blubber (57.53%). Oleic acid was the monene of highest concentration in all tissues examined. PUFA content was dependent on the tissue type and was in the order of blubber (21.27%), muscle (24.65%), brain (26.24%), kidney (26.79%), heart (30.19%), and liver (33.98%).

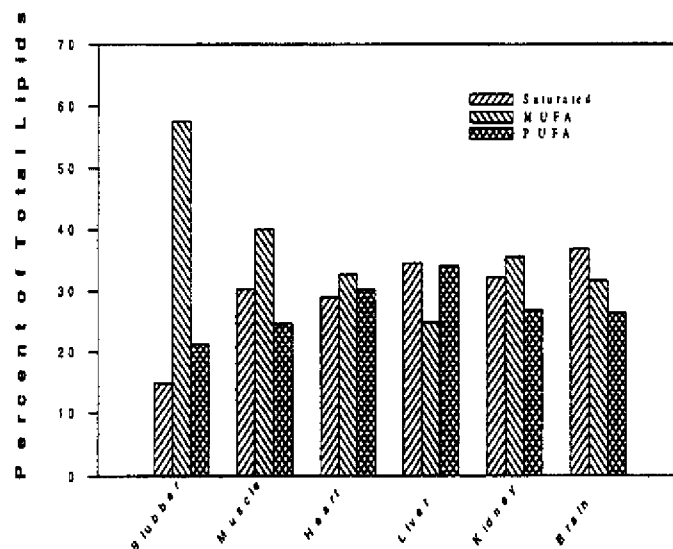


Figure 2. Content of saturated, monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids in selected tissues of grey seal.

The content of individual omega-3 PUFAs in each selected tissue is given in Figure 3. These were also dependent on the tissue type with brain lipids having the highest content of DHA and the lowest proportion of EPA. DPA was most abundant in the blubber and lowest in the heart lipids. Meanwhile, DHA was lowest in kidney lipids, while liver contained the highest levels of EPA.

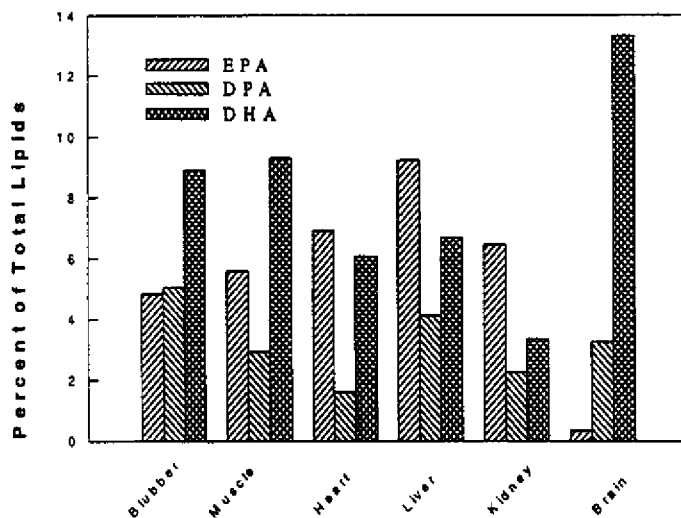


Figure 3. Eicosapentaenoic (EPA), docosapentaenoic (DPA), and docosahexaenoic (DHA) acid contents in selected tissues of grey seal.

Lipid Classes

Lipid classes of different tissues of grey seal, as determined by the TLC/FID, indicated that most tissues had appreciable amounts of triacylglycerol, cholesterol, phosphatidylcholine, phosphatidylethanolamine and phosphatidylserine. Blubber was primarily composed of triacylglycerols (99.3%), while brain lipids had no detectable amounts of triacylglycerol. Brain lipids had the highest level of cholesterol. Phosphatidylcholine had the highest content of polar lipids in all tissues except brain. Cerebroside was the dominant polar lipid constituent in the brain of grey seals.

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ABSTRACTS

SENSORY EVALUATION AND TEMPERATURE ABUSE STUDY OF "AMERIPURE" PROCESSED OYSTER

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"AmeriPure" process has proven to be effective in reducing the health risk associated with *Vibrio vulnificus* in the shellstock oyster to undetectable level and maintain the microbial quality of oysters following the AmeriPure process. Oysters were evaluated by a taste panel consisting of 10 LSU students and faculty. The sensory attributes evaluated included shape, off odor, off flavor, good oyster flavor, texture and overall acceptability. Both control and AmeriPure processed oysters by two different packaging methods were evaluated in three replicates from September to December, 1955.

"AmeriPure" process significantly slowed the rate of the production of off odor and off flavor in oysters during ice storage when compared to the control samples. In addition, the "AmeriPure" process slowed oysters shrinkage while enhancing good oyster flavor. The results of this sensory evaluation showed that "AmeriPure" process didn't significantly alter the sensory quality of oyster and extended the shelf life of processed oyster at least 7 days the control sample.

At all stages of post process, storage, transportation and "point of sale", oysters may be subjected to temperature abuse. This temperature abuse study analyzed the microbial quality of the "AmeriPure" processed oysters which were exposed to ambient temperature of $22 \pm 2^\circ\text{C}$ for up to 24 hours. The microbiological analyses showed no recovery of *Vibrio vulnificus* after 24 hours of temperature abuse. However, other aerobic bacteria showed a rapid growth after 10 hours of exposure and oysters were considered spoiled by 24 hours of exposure.

MICROBIAL LOAD AND PROFILE OF CHANNEL CATFISH FILLETS

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Channel catfish (*Ictalurus punctatus*) filets harvested in the fall of 1995 were processed by five different ways; manual deheaded-maulaul fillet/skin-chill-fillet (mDmF/SCF), manual dehead-automatic fillet not chilled (MdaF/UC), automatic dehead-automatic fillet/dehead-chill-fillet (aDaF/DCF), manual dehead-automatic fillet/eviscerate-chill-fillet (mDaF/ECF), and automatic dehead-automatic fillet/trim-chill-pack (aDaF/TCP). They were examined for microbial profile, psychotropic (PPC) and total coliform (TCC) counts. The predominant bacteria genotypes found in larger proportions in product filleted and trimmed prior to chilling or packed not chilled isolated from all process flows were *Acinetobacter* (10.64%), *Flavobacterium* (8.51%), and *Pseudomonas* (7.09%). *Staphylococcus* (6.38%) were predominant in products filleted before skinned. However, there was no significant difference ($P>0.05$) among five different processing procedures on PPC and TCC in filets.

MICROBIAL LOADS IN CATFISH PROCESS MACHINERY, PRODUCTS AND ENVIRONMENT DURING ONE DAY OPERATIONS

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Aerobic (APC), psychotropic (PPC), and total coliform (TCC) counts were determined in three different channel catfish operations. About 60 different sampling points per operation were monitored hourly for one day (8 hours). Product from manual and automatic filleting lines, breading and marinating line, machinery and conveyor belts, and environmental points were sampled. APC of fish skin and viscera were 2.8-5.0 log CFU/cm² and 3.0-7.1 log CFU/g, respectively. TCC for the same were 2.0-3.0 and 2.0-5.4 log CFU/cm². These are thought to be the points of highest microbial load. Dressed fish AFP were 3.0-5.8, nuggets had 3.0-4.8, filets had 3.3-6.5, and breaded nuggets had 3.0 log CFU/cm². Conveyor belts had APC ranging from 3.0-7.4 and TCC from 1.8 to 3.8 log CFU/cm². Holding vat water, filleter's knife, trimming board, and phosphate mix were found to have high APC, PPC and TCC. Cleaning, sanitation, and rapid cooling/chilling of products were found to be the main sources of contamination, in addition to fish skin and viscera.

STORAGE CHARACTERISTICS OF PASTEURIZED BLUE CRAB MEAT AND CRYOGENICALLY AND BLAST FROZEN CRAB MEAT HELD IN VACUUM AND MODIFIED ATMOSPHERE PACKAGES

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Cryogenic and blast freezing of special blue crab meat held in improved packaging materials were investigated. The experimental packages were: (1) vacuum packaged Cryovac boil-in-bags, (2) Dynopack trays, and (3) Dynopack trays with a nitrogen atmosphere. Packaged meat was either frozen in a commercial CO₂ tunnel (-54°C) or in a commercial blast freezer (-19°C). Crab meat pasteurized in steel-tin cans served as the control. Frozen crab meat was transferred from the blast freezer to a walk-in freezer and held at -12°C for ten months. Pasteurized meat was kept on ice in a walk-in cooler. The following analyses were completed at 1, 2, 4, 6, 8, 10, 12, and 15 months of storage: headspace O₂ and CO₂, NH₃, pH, aerobic plate counts, percent moisture, and Hunter L, a, b color values. A six-member trained panel developed sensory appearance, odor, taste, texture, color, general appearance, and texture attributes.

The following conclusions were drawn from the study: (1) freezing crab meat produces less bluing than pasteurizing crab meat, (2) although pasteurized crab meat is bluer than frozen meat, it has a whiter component than frozen meat, (3) the sensory panel found no consistent textural differences between frozen and pasteurized meat, (4) aerobic plate counts correlated well with storage time for pasteurized meat, but not frozen meat, (5) the microbiological shelf life of crab meat pasteurized according to National Blue Crab Industry Association Guidelines was exceeded after 12 months of refrigerated storage, (6) frozen shelf life was maintained through 15 months, (7) CO₂ freezing at -54°C did not significantly improve crab meat quality attributes when compared to blast freezing at -19°C, and (8) properly frozen and stored blue crab meat may provide a commercial alternative to hot water pasteurization.

PHYSICOCHEMICAL AND SENSORY CHANGES IN FROZEN HYBRID-STRIPED BASS FILLETS

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Hybrid-striped bass, HSB (*Morons saxatilis x Morone chrysops*), were treated by a single glaze, double glaze (water), dipped in erythorbic acid, and in Tenox 20A®. They were stored at -18°C for five months and analyzed for carbonyls, 2-Thiobarbituric acid reactive substances (TBARs), Hunter color, and sensory ratings. Carbonyls were lower in fillets treated with antioxidants as compared to glazed fillets, over frozen storage. TSARs of raw and cooked products did not exceed 1.50 for all treatments. Sensory flavor ratings ranged from two to over four, but were not influenced by treatment. Hunter 'L' values were apparently higher in Tanox 20A® treated fillets while 'a' values were lower. There was little difference in fatty acid profiles between raw and baked fillets. Data shows that HSB frozen fillets are very stable for five months of frozen storage regardless of glaze or antioxidant treatment.

CRYOPROTECTIVE MECHANISM OF MALTODEXTRIN IN FROZEN SURIMI

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We investigated the cryoprotective properties of maltodextrins in surimi, and additionally sought to determine whether high molecular-weight carbohydrates differ in their cryoprotective mechanism from low molecular-weight compounds. A range of maltodextrins (DE 4-25) were added to Alaska Pollock surimi at 8% and tested against sucrose and sucrose/sorbitol controls. The surimi was quick frozen, then later equilibrated to storage temperatures of -20°C, -14°C and -81°C at 6 months intervals. Additionally these samples were subjected to freeze/thaw cycling. Protein quality at time of sampling was determined by measuring CA⁺ATPase activity. Surface tension of maltodextrin solution was also measured.

Cryoprotectancy of the maltodextrin exhibited a slight decline in the remaining enzyme activity as MW of the cryoprotectant increased, an effect that was more pronounced at the higher storage temperature. In contrast, for samples subjected to F/T cycling the residual activity was reduced as the MW of the cryoprotectants increased. This evidence supports a dual mechanism for cryoprotection by carbohydrates based on their molecular weight. Lower MW maltodextrins enhance surface tension of the solutions and they may cryoprotect by the same mechanism (preferential exclusion) as sucrose. Higher MW maltodextrins were significantly effective cryoprotectants at storage temperatures near or below their respective Tg' values but ineffective protectants in F/T cycling and they did not increase the surface tension.

PRODUCTION AND CHARACTERISTICS OF FLAVORING FROM SQUID BY-PRODUCT HYDROLYSATE

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An orthogonal array at $L_4(2^3)$ was used to determine an optimal condition for the production of flavoring from cooked or uncooked squid by-products by enzymatic hydrolysis and spray drying using Flavorzyme and Protamex (Novo Nordisk Blochem). The most favorable flavor profile was obtained from uncooked squid after 6 hr hydrolysis at 50°C followed by 2 hr maturation. Addition of spray-dried flavoring increased the taste intensity of surimi-based squid analog. Predominant free amino acids liberated during hydrolysis were arginine, leucine, alanine, lysine, glutamic acid, and glycine, which are believed to contribute to the squid flavor.

STATUS OF THE CUBAN SEAFOOD INDUSTRY AND IMPLICATIONS FOR FLORIDA OF RENEWED TRADE

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The Cuban commercial fishing industry existed primarily as an artisanal fishery until the late 1950s. Following the Revolution and the subsequent US embargo, the Cuban commercial fishing industry expanded in capacity and technological sophistication through Soviet subsidization. This led to the development of a large distant-water fleet and a revitalized nearshore fleet. With the cessation of Soviet subsidization in 1991, however, the Cuban commercial fishing industry is undergoing radical changes in capacity utilization and management structure. Foreign investment is being encouraged as a means to finance the renovation of existing processing facilities. Current production is focusing on high-value species, which are being retained in greater volumes within the domestic market. However, renewed trade with the US would allow significant quantities to enter the Florida seafood market, with differential impacts to the Florida harvesting and processing sectors.

HACCP IMPLICATIONS OF MICROBIAL LEVELS IN SEAFOOD BATTER OPERATIONS

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Batter operations were monitored during full-scale production of breaded raw fish portions, breaded raw butterfly, and breaded precooked popcorn shrimp. Microbial loads in fish batter were APC $5.53\% \times 10^3$ CFU/g and coliform 4.0×10^2 CFU/g. Microbial loads in fish batters were APC 6.97×10^2 CFU/g and *S. aureus* 6.62×10^1 CFU/g. Microbial loads in popcorn shrimp batter were slightly higher with APC 9.85×10^3 CFU/g, coliform 5.93×10^2 CFU/g, *E. coli* 8.61×10^1 CFU/g, and *S. aureus* 3.48×10^2 CFU/g. The implications of these data in relation to the significance of time and temperature abuse and monitoring requirements under the U.S. mandatory HACCP inspection program will be discussed.

IMPORTANT CONSIDERATIONS FOR THE QUALITY AND WORKMANSHIP OF SEAFOOD PRODUCTS

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There is a tremendous increase in focus on HACCP within the seafood industry. Many people within the industry are involved in conducting training classes on getting trained. Others are involved in the purchase of new or additional equipment and services that will assist them in establishing critical limits, monitoring and verification. All of this is important and essential to minimize the risk of chemical, biological and physical hazards and contamination in seafood products for human consumption.

It is also important for us to not forget about those factors and product characteristics that affect the eating quality, aesthetics and performance of the product by the end user. Product texture, flavor, uniformity of size, neatness of cut, proper trimming and odor, are a few of the characteristics that impact seafood quality and sales.

SEAFOOD QUALITY AND SAFETY MANAGEMENT SYSTEMS IN RETAIL SEAFOOD DEPARTMENTS

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The current state of product quality, shrinkage and safety management programs was studied in retail seafood departments. Eleven stores from four major U.S. retail chains in the South and Mid-Atlantic were audited for three days each. Among the parameters documented were 1) product handling methods. Selected stores also received a microbiological audit of environmental surfaces or products. Laboratory studies were conducted to determine the effect of display methods on product weight loss and seafood salad shelf-life (sensory, APC). Results indicate that seafood department facilities and equipment are generally adequate and properly controlled. However, procedures related to sanitation, cooking and management of ice-only display cases were highly variable. APCs of fresh flounder fillets were high ($x=10^7$ cfu/g). No pathogens were isolated from ready-to-eat products. Shrinkage rates were product specific. Product quality and potential safety were most frequently compromised by cross-contamination events related to a lack of chain-wide operating procedures. A model SOP is proposed for in-store cooking.

DESIGNING STANDARD OPERATING PROCEDURES FOR FULL SERVICE RETAIL SEAFOOD DEPARTMENTS IN SUPERMARKETS

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Grocers have added features which provide shoppers with many of the conveniences found in food service establishments. These include: full service, more emphasis on perishables, and more refrigerated, ready-to-eat items which are prepared on site. Most full service seafood departments also embody these features.

Such additions have resulted in more complex retail operations. This suggests the need for experienced, technically proficient labor pool. However, success in food retailing depends upon cost control. Therefore, technical competence is generally traded off for the cost savings that part time positions provide. Unfortunately, maximize shelf while minimizing product safety threats.

While the goals necessary to minimize shrinkage and compromised product safety are common knowledge, translating them into operational procedures has been a difficult undertaking for the food retailing community. This project explored root causes for quality and safety errors, and built research-based Standard Operating Procedures which are effective, simple, and time efficient to implement by the current labor force.

QUICK TIPS FOR FOOD HANDLERS

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-- No abstract submitted --

PRELIMINARY OBSERVATIONS ON A HAPLOSPORIDAN PARASITE IN THE CATARINA SCALLOP *ARGOPECTEN CIRCULARIS* HARVESTED FROM BAJA CALIFORNIA SUR, MEXICO

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The Mexican bay scallop *Argopecten circularis*, commonly referred to as the Catarina scallop, is a subtropical species commercially harvested around the Peninsula de Baja California in bays along the Northwest Golfo de California and the Pacific coast. The scallops are hand harvested and shucked at the harvesting sight, retaining only the adductor muscle. The shucked meats are iced on trucks, transported fresh to the U.S. and distributed by the importer either fresh or frozen. Upon evaluating a large volume of frozen Catarina scallop meats, an estimated 1-2% of the meats were macroscopically observed to have brown or black cysts with intermuscular orientation both on the meat surface and embedded deep within the meat. The scallop meats affected either had all brown spots, or all black spots, with only a few meats observed to possess both. The cysts differed in that the brownish cysts had soft cyst walls each containing puss-like degenerative tissue, while the black cysts possessed somewhat hardened, fibrous cyst walls and contained numerous parasitic protozoan spores. Upon analyzing morphological characteristics of the spores utilizing histological (H&E staining), and SEM and TEM techniques, the spore were preliminarily identified as *Urosporidium* ssp. within the Phylum Haplosporidia. The spores were observed to be grouped within numerous membrane-bound sporocysts within each cyst. Major spore morphological characteristics include a mostly spherical spore (average diameter 7.75 microns, n=9) possessing an internal flap or lingula for closure of the spore orifice, and a highly flexible, singularly appearing extension (tail) averaging 39.6 microns in length (n=9) extending in a posterior direction in relation to the spore orifice. The tail is observed to be made up of numerous long extensions of the spore wall, which intertwine in a twisting arrangement as they extend distally.

The presence of this parasite may or may not be of commercial importance. *Urosporidium crescens*, the hyperparasite of the parasitic microphalli fluke found in blue crab muscle, has indirectly been of some commercial significance along the US east and Gulf Coasts.

AUTOMATION OF QUALITY EVALUATION OF SALMON: A PRELIMINARY STUDY

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Current inspection of salmon quality relies on sensory evaluation by inspectors who evaluate the fish for visual, smell and texture attributes. This procedure is subjective, prone to error, time consuming, difficult to relate to others, and to quantify. This study used a combination of machine vision analysis and electronic nose technology to correlate objective attributes of salmon to sensory evaluations from a panel.

Atlantic salmon (*Salmo salar*) filets from Chile was obtained fresh, within 48 hrs of harvest. The fish were stored at 35, 45 and 55°F for up to 10 days. Samples were also stored in variable temperature environments. Each day, an image of the flesh and the skin sides were captured, and the fish were analyzed by Neotronics electronic nose. Six replicates were performed per analysis. A sensory panel also evaluated the fish for visual and smell attributes. It was found that two colors could be correlated with storage time and sensory grade. Predictive equations of color change with time were also developed. These results could be used to develop methodologies to assist in the objective and repeatable quality evaluation of salmon.

COOKING OF TIGER SHRIMP: MODELING QUALITY AND SAFETY

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The quality of thermally processed shrimp is determined by microbiological and textural attributes. Yield loss is another important factor. These attributes are related to the product's thermal history and temperature distribution. Since it is impossible to monitor the temperature at all points in the shrimp, a mathematical model was developed for the prediction of its temperature distribution during cooking and cooling. Three sizes of tiger shrimp (large, 16-20/lb; medium, 41-50/lb; small, 50-60/lb) were cooked in water at 75, 85, 95 and 100 °C for different periods. Cooling was done in icewater, or in ziploc bags in icewater, to observe effects of cooling methods on the yield loss and texture parameters. After each treatment, the yield loss and changes in texture parameters were measured. For large tiger shrimp, percent yield loss was 10% when cooked at 75 °C for 5.6 min, and 25% at 100 °C for 2.6 minutes. Safety was determined by selecting *Vibrio cholera* as the target microorganism. A 6 log cycle reduction at the slowest heating point of shrimp was achieved for all tests. Texture profile analysis parameters were measured using an Instron Machine, and the sensory parameters of toughness, juiciness, rubberiness and overall acceptability were determined by taste panel tests. Correlation of these parameters was accomplished.

The mathematical model can predict the effect of cooking and cooling on the yield loss and shrinkage, on texture, and on the desired safety of the shrimp. This tool can be used to optimize the cooking parameters of tiger shrimp.

**FLORIDA'S APPROACH TO THE INTERIM CONTROL PLAN (ICP) FOR
REDUCING THE RISK OF *VIBRIO VULNIFICUS* INFECTION
AND THE USE OF TIME-TEMPERATURE
RECORDERS AND INTEGRATOR**

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Sensitech, Inc. and *VITSAB

As a response to the consistent number of *Vibrio vulnificus* (V.v.) infection cases in the United States the Interstate Shellfish Sanitation Conference (ISSC) has adopted an Interim Control Plan (ICP) (August 25, 1995). This ICP has the objective of retarding the growth in numbers of V.v. in Gulf of Mexico Oysters (*Crassostrea virginica*) to be consumed raw on the half-shell. The ICP classifies the harvesting periods into four levels depending on the water temperature with each level having a specific time-temperature matrix intended to limit V.v. densities after harvesting.

The State of Florida, as of May 1996, adopted the matrix in order to be consistent with the ISSC in the prevention of V.v. infection. Even though all shellstock harvested from Florida's waters already complies with the matrix the FL Department of Environmental Protection has agreed to cooperate in the validation of this time-temperature regime. Starting the month of November and continuing throughout a period of 12 months samples will be collected and analyzed for V.v. bacteria. This study will compare the matrix handled product against a non matrix product. In addition the State of Florida has joined forces with the University of Florida as well as with two companies producing time-temperature integrators and recorders to help follow the product regime through harvest and production. The intent is to investigate the utility of a series of time-temperature integrators (VITSAB's) that can be activated on site and change colors progressively to indicate the time the oyster must be placed into a mechanically refrigerated room. The recorders or electronic data loggers (TempTales - Sensitech) will monitor actual time-temperature consequence during all handling and storage.

Florida is a major player in shellstock production and must always be on the cutting edge looking for better and safer ways to process the oysters to assure confidence to the consumers.

**THE EFFECTS OF SODIUM LACTATE ON COLD SMOKED SALMON IN
TERMS OF BACTERIAL REDUCTION, INCLUDING *LISTERIA*
*MONOCYTOGENES***

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Cold smoked salmon has been documented to contain a variety of pathogenic bacteria and viruses (Noah, 1991; Jemmi, 1990; Weagant, 1988). In recent years, the smoked salmon industry has received a great deal of attention due to *L. monocytogenes* contamination. This pathogen has been associated with several outbreaks in North America and Europe (Bille, 1988; Buchanan, 1989; Fleming 1985). Listeriosis occurs mainly in pregnant women, their unborn children, and immunodeficient individuals (Fleming 1985). The United States Food and Drug Administration has adopted a "zero tolerance" policy for *L. monocytogenes*. This emerging foodborne pathogen has been isolated in raw and ready-to-eat fishery products (Buchanan 1989). Jemmi (1990, 1994), and Fuchs (1994) isolated *L. monocytogenes* from wholesale and retail smoked salmon products in 24.0, 30.8 and 8.6 percent of analyzed samples, respectively. Little data is available on the incidence and behavior of *L. monocytogenes* during preparation, processing and storage of smoked salmon.

Fresh raw salmon fillets were subjected to different treatments of sodium lactate (NaL) during brining and subsequently analyzed microbiologically and chemically. Salmon fillets were brined for 18 hrs or 7 days with the addition on various concentrations of NaL (0.0, 1.24, and 2.48%). This investigation also addressed the effects of a topical 5.0% sodium lactate wash on fillets inoculated with a genetically engineered bioluminescent *L. monocytogenes*. The salmon fillets were stored under vacuum in low oxygen permeable packaging at $-10 \pm 1^{\circ}\text{C}$ and enumerated for *L. monocytogenes* and total aerobic microorganisms at specific intervals up to 30 days. In addition, pH, water activity, and sensory characteristics in processed smoked salmon were also examined.

Results from this study indicate that sodium lactate was limited in its' ability to inhibit or delay the growth of either naturally occurring or artificially inoculated salmon with *L. monocytogenes* during both the brining and/or cold smoking process. However, the antimicrobial effects of the NaL were very effective on total aerobic plate counts. A 5.0% NaL wash was also proven to be effective in reducing aerobic microflora in both the brining and cold smoked salmon. Sodium lactate had no affect on pH, a_w , or color of the cold smoked salmon vacuum packaged and stored at -10°C for 30 days. The taste and texture of NaL treated fillets were reported to be more desirable than those of the control group.

