

Development of a HACCP-based Strategy for the Control of Histamine for the Fresh Tuna Industry.

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TABLE OF CONTENTS

	<u>Page</u>
I. Report Title, Author, Organization, Grant Number and Date	2
II. Abstract: A brief (one paragraph) description of the final report (for use in the S-K Annotated Bibliography).	2
III. Executive Summary: A brief succinct summary of final report.	3
IV. Purpose:	4
A. Detailed description of problem or impediment of fishing industry that was addressed.	4
B. Objectives of the project.	7
V. Approach:	8
A. Detailed description of work that was performed.	8
B. Project Management: List of individuals and/or organizations actually performing the work and how it was done.	11
VI. Findings:	12
A. Actual accomplishments and findings.	12
Results Objective 1.	12
Results Objective 2.	14
Results Objective 3.	18
Results Objective 4.	31
Results Objective 5.	32
Results Objective 6.	40
Results Objective 7.	44
B. If significant problems developed which resulted in less than satisfactory results, they should be addressed.	44
C. Description of need, if any, for additional work.	45
V. Evaluation:	46
A. Describe:	46
1. Were the goals and objectives attained?	46
VI. References:	47

Final Report

I. Report Title, Author, Organization, Grant Number and Date

Title: Development of a HACCP-based strategy for the control of histamine for the fresh tuna industry.

Organization: PacMar, Inc., Honolulu, Hawaii

Grant No. NA86FD0067

Date: July 31, 2000

II. Abstract: A brief (one paragraph) description of the final report (for use in the S-K Annotated Bibliography).

Histamine or scombroid fish poisoning is among the top three seafood-related public health problems reported in the US. Epidemiological data from Hawaii between September 1989 and September 1999 indicate that mahimahi (54%) and tuna (25%) were the leading fish species implicated in illnesses due to histamine poisoning. Imported seafood was responsible for 48% and imported mahimahi was responsible for 45% of the total number of illnesses. A practical HACCP-based approach (Hazard Analysis Critical Control Point) for controlling histamine accumulation in susceptible pelagic fish species caught by Hawaii's longline, handline and trolling fleets was explored. The FDA HACCP seafood inspection program guidelines for controlling histamine accumulation recommend that fish be chilled to below 50° F within 6 hours and to below 40° F within 24 hours after death. Vessel Standard Operating Procedures (VSOP) for on-board fish handling were evaluated against these guidelines. Fish temperature profiles were recorded at sea and compared with histamine analyses. Hawaii fishing fleets were capable of meeting the FDA fish handling guidelines for fish brought to the vessel alive. The actual chilling rates for fish that died on the line could not be determined, however, once boarded, fish were chilled to below 40° F within 24 hours. The histamine concentration of all fish (dead and alive) with known on-board temperature profiles was well below the FDA defect action limit of 5 mg/100g (mean = 0.26 mg/100g, range = 0.02 – 0.88mg/100g, SD 0.2 mg/100g). The efficacy of fish quality grading and sensory evaluation at the time of delivery to the first receiver was evaluated as a practical screening method for eliminating fish with high histamine risk from the market. A market sample of 583 fish from 42 commercial longline trips, 45 trolling trips and 32 handline trips was collected, graded for quality, evaluated organoleptically and analyzed for histamine concentration. Fish quality grading and sensory evaluation (for odors of decomposition) were effective in culling out all fish (14 out of 583 fish sampled) with high histamine concentrations. The fish rejected for odors of decomposition included, bigeye, yellowfin and albacore tuna, striped marlin, blue marlin and mahimahi. Within the sample set of odor rejects, only bigeye tuna, yellowfin tuna, albacore tuna and mahimahi were found with histamine levels exceeding the defect action limit. It was estimated that the actual prevalence of high histamine fish in Hawaii's fresh fish landings is less than 0.00117%. A practical HACCP-based approach utilizing VSOP for controlling histamine on fishing vessels and sensory evaluation for screening for fish with high histamine risk in the Hawaii fresh tuna industry is presented.

III. Executive Summary: A brief succinct summary of final report.

A study was conducted to determine how histamine forms during the post-harvest period on-board commercial fishing vessels in Hawaii's fresh tuna industry. A practical and effective, HACCP-based approach to histamine control was developed.

The epidemiological records for outbreaks of histamine poisoning in Hawaii between September 1989 and September 1999 were evaluated. Tuna and mahimahi were the two most important species being implicated in 68% of the histamine incidents and 80% of the number of illnesses.

Further analysis revealed that imported fish caused 48% of the histamine illnesses. Imported mahimahi caused 45% of the histamine illnesses.

The on-board fish handling methods used by Hawaii's commercial troll, handline and longline fleets were monitored and documented at sea using temperature loggers to accurately profile the time and temperature parameters in the post-harvest period.

On-board fish handling methods were compared with FDA fish handling guidelines for the prevention of histamine accumulation. The commercial vessels studied were capable of meeting the guidelines with fish brought aboard alive. For fish brought aboard dead, the actual chilling times (from the time of death) could not be determined.

Histamine analysis was conducted to verify that the handling methods observed adequately controlled histamine accumulation. All fish, dead and alive, with known on-board temperature profiles were well below the FDA defect action limit of 5 mg/100g (mean = 0.26 mg/100g, range = 0.02 – 0.88 mg/100g, SD 0.2 mg/100g) at unloading.

The efficacy of fish quality grading and sensory evaluation of fish for odors of decomposition as a practical means of culling fish with high histamine was evaluated by sampling 583 fish in the Hawaii fresh fish landings. Fish from troll, handline and longline vessels were sampled. Bigeye tuna, yellowfin tuna, albacore tuna, striped marlin, blue marlin and mahimahi were sampled. The sampling protocol called for collecting near equal numbers of fish from each of the 5 grade categories. Grade 5 fish (odor rejects) were rare, however eventually 119 odor rejects were collected for the study. All fish were sampled and analyzed for muscle histamine concentration.

A total of 14 fish out of the entire market sample of 583 fish were found to exceed the histamine defect action level of 5 mg/100 g. All of these fish were first rejected from the market for odors of decomposition (Grade 5).

It was concluded that odors of decomposition are reliable indicators of histamine risk and that sensory evaluation is an effective HACCP control measure in the Hawaii fishery.

It was estimated that the actual prevalence of high histamine fish in Hawaii's fresh fish landings is less than 0.00117%.

A practical HACCP-based system for histamine control in the Hawaii fresh tuna industry was developed that integrates new information generated by the study on the efficacy of VSOP in controlling histamine accumulation and the efficacy of sensory evaluation in screening for high histamine risk fish.

IV. Purpose:

A. Detailed description of problem or impediment of fishing industry that was addressed.

Histamine poisoning is the most important seafood-related public health problem facing the US fresh tuna industry today. It is caused by the consumption of fish containing toxic concentrations of histamine and other biogenic amines (Taylor et al., 1984). Histamine poisoning is also known as "*scombroid fish poisoning*" although non-scombroid fish species are often involved. Mahimahi, tuna and bluefish are the fish most commonly implicated in the US.

Histamine accumulates to toxic levels in certain fish species that have naturally high levels of the free amino acid, histidine. If these fish are subjected to significant mishandling in the post-harvest period (prolonged temperature abuse and poor sanitation), bacteria proliferate. Certain species of bacteria are histamine formers, capable of producing the enzyme, histidine decarboxylase. This enzyme converts free histidine into histamine, the predominant toxin responsible for scombroid fish poisoning. If these histamine-forming bacteria species are present, toxic levels of histamine may accumulate.

The US Food and Drug Administration (FDA) is overseeing the program of mandatory food safety controls in the seafood industry. The FDA adopted HACCP (Hazard Analysis Critical Control Point) as the basis for its inspection program that became effective in December of 1997. The HACCP principle is based on the premise that the prevention of hazards is vastly more effective than end-product sampling and testing as a means of controlling seafood safety hazards. This program requires that all seafood processors conduct a hazard analysis of their products and processes, draft a plan for monitoring critical control points in the process, establish a system for record-keeping and prepare plans for corrective actions when critical limits are exceeded.

HACCP is a process control philosophy that relies on monitoring critical control points in the process to prevent food safety hazards that are deemed "*likely to occur*" based on the best available scientific and industry knowledge. HACCP is not a zero-risk system, but is aimed at limiting likely hazards. FDA HACCP is also not a "*prescriptive program*" that dictates exactly how companies or industry sectors choose to control recognized food safety hazards. Rather, the industry is encouraged to apply practical knowledge in combination with scientifically valid applied research and guidance to develop practical and effective food safety controls tailored to particular products, processes, facilities and industry sectors.

Histamine poisoning is a self-limiting, pseudo-allergic reaction to the consumption of fish containing toxic levels of histamine, a mediator of the immune response. It is easily treated with anti-histamines and although it causes great alarm and discomfort, it is not considered a lethal hazard. However, histamine poisoning is one of the most important seafood-related public health issues being addressed in the nationwide FDA HACCP program, considering the number of reported illnesses, the range of species implicated, the product forms and volume of histamine-susceptible seafood consumed in the US.

The FDA suggests two basic alternatives for first receivers (referred to as "primary processors" by the FDA) for controlling histamine in fresh tuna and associated pelagic species received from the primary producers (fishing vessels).

The Harvest Vessel Controls Approach.

The first method relies on receiving detailed on-board fish handling monitoring records from the vessels that document when fish were caught and details of the temperature history of the fish during the initial cooling period and subsequent storage phase. The FDA has developed fish handling guidelines (Table 1) for time and temperature controls needed to prevent excessive histamine accumulation (FDA, 1998).

Table 1. FDA fish handling guidelines for the prevention of histamine accumulation.

Category of histamine-producing fish.	On-board Fish Handling Guidelines.
For fish other than tuna above 20 lbs., if the fish has not been exposed to temperatures above 83° F (28.3° C),	The fish should be placed in seawater or brine at 50° F (10° C) or less within 9 hours of death. OR The fish should be placed in ice within 12 hours of death.
For tuna greater than 20 lbs., OR If the fish have been exposed to temperatures above 83° F (28.3° C),	The internal temperature of the fish should be brought to below 50° F (10° C) within 6 hours of death.

Primary processors using the Harvest Vessel Approach shall at the time of delivery,

- Receive detailed fish handling records from the vessel operator.
- Conduct sensory evaluation for decomposition by sampling 118 fish in a lot (or each fish for lots <118 fish) and reject entire lot if rejects exceed 2.5% or 3 fish.
- Check the adequacy of the icing for other cooling media.
- Record the internal body temperature of the fish at the time of delivery.

The Histamine Testing Approach.

The second alternative relies on lot sampling and testing of fish for histamine. The FDA policy for histamine concentration in the edible portion of fish is given in Table 2.

Table 2. FDA policy for histamine concentration in seafood product.

Histamine Concentration	FDA Policy
5 mg/100 g (50 ppm)	Defect Action Limit
50 mg/100 g (500 ppm)	Estimated toxicity level

The FDA recommends that the primary processor collect muscle samples for histamine analysis from 1 fish per ton for large fish (>20lbs.) and 2 fish per ton for smaller fish (<20lbs.) where the fish are from common origin (FDA, 1998). The lot sampling and testing of fish landings is not based on HACCP, does not emphasize prevention of the hazard through processing controls and is a form of end product testing. Lot sampling and testing may not be an effective method for detecting histamine in fresh fish from

hook and line fisheries because of the way fish are caught and handled in the post-harvest period.

Primary processors using the Histamine Testing Approach, shall at the time of delivery,

- Lot sample and test for histamine.
- Conduct sensory evaluation for decomposition by sampling 118 fish in a lot (or each fish for lots <118 fish) and reject entire lot if rejects exceed 2.5% or 3 fish.
- Check the adequacy of the icing or other cooling media.
- Record the internal body temperature of the fish at the time of delivery.

For many years the tuna canning industry has relied on a Histamine Testing Approach to screen frozen tuna for elevated histamine concentration. The sampling frequency is 1 fish per ton for fish greater than 20 lb round weight, and 2 fish per ton for fish that are less than 20 lb. In the western Pacific purse seine fishery, many of the fish that are caught are less than 20 lb. Lot size may be as large as 90 tons with the assumption being that each fish well should be considered a single lot. Fish wells on US purse seiners hold from 20 to 90 tons of fish. In this fishery individual sets can exceed 400 tons of fish and fill multiple fish wells. Fish are loaded into the well, usually from the same purse seine set, chilled in refrigerated seawater, frozen in brine and stored frozen. With 5 lb fish for example, this is equivalent to 400 fish per ton or up to 36,000 individual fish per 90-ton lot. This amounts to a sampling frequency of 0.25%. This low sampling rate is only likely to be effective in situations where there has been gross mishandling of the fish resulting a high percentage of fish with elevated histamine in the lot.

The sampling rate is not sufficient to detect the few individual fish with high histamine or decomposition that may occur in a lot at extremely low frequency. Because of this Quality Control personnel at the canneries detect and cull individual decomposed fish from production lines by having "sniffers" screen each fish entering the pre-cooking stage. This step in combination with lot sampling and testing for histamine is effective in reducing the histamine risk and makes canned tuna a very safe product.

In contrast, the way in which fresh tuna are caught (hook and line gear), graded for quality and sold on individual quality merits, makes the application of representative lot sampling ineffective. These fish are caught individually and not in large sets as with purse seines. Even within a single longline set, the series of fish caught have unique histories from the time they were hooked until they were placed into the hold. Each fish has its own time and temperature history at the end of the trip and there is no reason to consider all fish from the trip or each longline set a "lot". For this reason, the Histamine Testing Approach of lot sampling is of questionable value when applied to the fresh tuna industry.

Measures to control histamine accumulation in the fresh tuna industry should instead focus on prevention by applying time and temperature controls and sanitation procedures on vessels and continuing through the processing and distribution channels. The Harvest Vessel Approach relies on detailed on-board handling records from fishers that may be impractical to collect and may actually be counterproductive to the objectives of good handling practices for vessels storing fresh fish in ice. Monitoring the temperature of a fish once it is placed in the ice requires removing the fish and inserting a thermometer into the edible muscle. This slows the chilling process, creates an entry for bacteria and may lead to localized decomposition of adjacent muscle.

The practical question is how to design a HACCP-based system that emphasizes prevention, establishes a set of standard operating procedures for fish handling on vessels using ice that can reliably prevent histamine accumulation and can reduce the likelihood of the histamine hazard. The responsibility for verifying proper on-board handling by fishers lies on the primary processor. How to document and verify that fish have been properly handled on-board fishing vessels is the practical challenge.

The project was aimed at the development and verification of a practical alternative, HACCP-based approach to controlling histamine risk in the fresh tuna industry. Essentially, the project conducted a histamine Hazard Analysis of the fresh tuna fishery in Hawaii. This is the first step in developing a HACCP-based program. Hazard Analysis identifies the likely food safety hazards, in this case the presence of fish containing high histamine concentration and the on-board handling conditions that allow histamine to accumulate to toxic levels. The project focused on integrating effective histamine control measures by the fishing vessels and verification methods for use by the primary processor. This initial transfer of responsibility from the vessel to the on-shore distribution chain is critical. The fish handling steps at sea are the most important in setting the quality and safety parameters. Without proper fish handling at sea, the control of histamine in subsequent steps is unlikely.

The alternative VSOP HACCP-based strategy relies on,

- Establishing a set of Vessel Standard Operating Procedures (VSOP).
- Verifying that the VSOP are able to meet FDA time and temperature guidelines.
- Determining that the VSOP are effective at preventing histamine accumulation.
- Ensuring that the VSOP are followed on fishing vessels.
- Verifying that sensory evaluation of fresh fish is a practical and effective critical control measure for histamine.

B. Objectives of the project.

- Objective 1. Evaluate epidemiological data on histamine poisoning in Hawaii.
- Objective 2. Develop Fleet on-board handling profiles (trollers, handliners and longliners).
- Objective 3. Verify the post-harvest fish handling procedures.
- Objective 4. Verify the relationship between post-harvest fish handling procedures and histamine accumulation.
- Objective 5. Determine the importance of fish quality grades and odors of decomposition as indicators of histamine concentration.
- Objective 6. Develop a HACCP-based strategy for the control of histamine for the fresh tuna industry.
- Objective 7. Communication of results to the FDA Office of Seafood.

V. Approach:

A. Detailed description of work that was performed.

Methods Obj. 1. Evaluate epidemiological data on histamine poisoning in Hawaii.

The Epidemiology Branch of the State of Hawaii Department of Health, was contacted in order to obtain 10 years of available data on cases of histamine poisoning in Hawaii between 9/20/89 and 9/28/99. Information requested included the number of outbreaks (incidents), number of cases (people made ill), the species of fish implicated, origin of the fish, product form and the market segment involved. Information was evaluated for completeness and accuracy in determining the relative risk of histamine accumulation within the different fish species, fishing methods, product forms, product origin and market segment. Epidemiological data from CDC (The Centers for Disease Control and Prevention) were also requested in order to determine if additional detail on outbreaks in Hawaii might be available from that authoritative source.

Methods Obj. 2. Develop Fleet on-board handling profiles (trollers, handliners and longliners).

Efforts were made to rapidly characterize the major fishing fleets in Hawaii (troll, handline and longline) specifically in terms of capture, fish handling and storage methods. Fishers were interviewed to determine the on-board handling procedures. Available literature was reviewed. Concise fleet profiles were prepared describing fishing methods and post-harvest fish handling methods which typify the standard practices on vessels representative of the various gear types. The type of fish caught and the associated quality issues were also described for each gear type.

Methods Obj. 3. Verify the post-harvest fish handling procedures.

The project team made research trips on commercial fishing vessels to observe, monitor and verify the fishing methods and fish handling procedures to complete the fleet profiles and VSOP. Participating fishing vessels were selected as representatives of the fish handling practices of the three major fishing gears.

The fishing methods were observed on trolling, handline and longline vessels during commercial fishing trips. Observations were recorded on how the fishing gear was used. For longline gear, the time at the start of the set and when the individual fish were hauled on-board was recorded. The fish species and condition (alive or dead) on retrieval were recorded. The weight of the fish (whole) was determined using a commercial platform scale (State of Hawaii-certified) at the time the fish were unloaded from the vessel and delivered to the first receiver in the market chain.

The fish handling methods were also observed and evaluated. How the fishers handled the fish immediately after being brought aboard was monitored. The processing time (deck time) from boarding to placement in the ice in the fish hold was recorded. The initial core fish temperatures were measured using a water-resistant microprocessor temperature meter (Hanna Instruments, HI 9024).

Continuous fish temperature profiles were recorded during the vessel phase of the post-harvest handling sequence using waterproof temperature loggers (Onset Computer Corporation, Stowaway® Tidbit XT). The stainless steel temperature probes (6-inch) were placed along the longitudinal axis of the vertebral column to record core muscle temperature (Figure 1). The temperature probes (TP) were placed into the muscle from within the gill cavity. The thermistor wires and the temperature loggers (TL) were then secured to the gill arches using cable tie wraps. Fish were identified with pectoral fin clips and heavy-duty plastic flagging ribbon around the caudal peduncle. Loggers were programmed to record temperature at various intervals (24 seconds to 5 minutes, depending on the length of trip) from the time the logger was triggered and placed into the fish until the logger was removed at the end of the trip during vessel unloading.

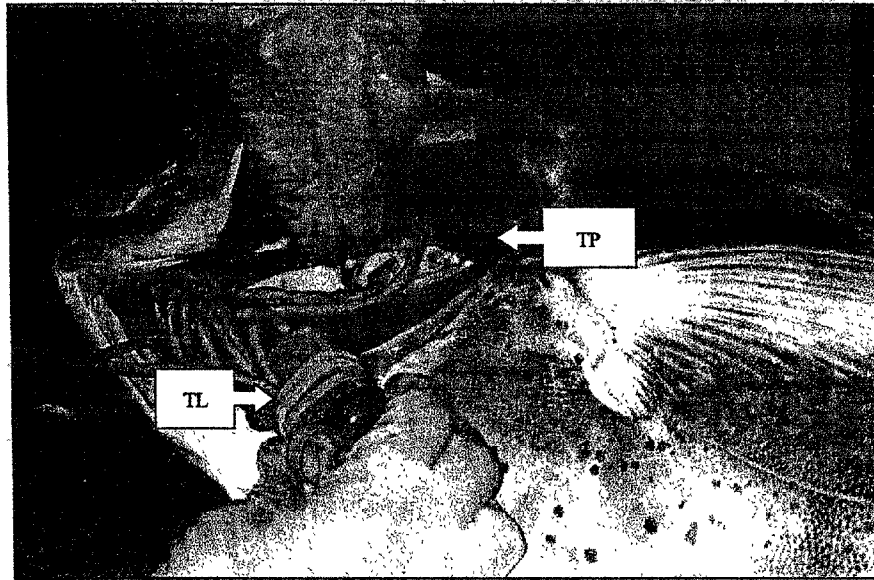


Figure 1. Placement of temperature logger (TL = temp. logger, TP = temp. probe).

Upon landing, the trial fish were identified as they were removed from the fish hold. The loggers were retrieved and the fish were weighed and evaluated organoleptically (sensory evaluation). The fish were also graded for quality using fresh tuna industry grading methods (Bartram et al. 1996). Quality grades used in the Hawaii fresh fish market were also applied to the associated pelagic fish species. A muscle sample was then collected from the dorsal muscle mass just posterior to the cleithrum for histamine analysis. Histamine is known to form earliest and reach the highest concentration in this anterior section of the body making it a logical sampling location (Frank et. al, 1981, Baranowski et. al. 1990). Muscle samples were immediately bagged, labeled, placed in ice and delivered to a freezer within 2 hours. Frozen samples were then delivered to the laboratory for histamine analysis.

The fish handling data collected during fishing trips were compiled and analyzed. The information relating to time and temperature targets for properly chilling fish was focused on the time it took fish to be chilled below 50° F, the time to reach below 40° F, and the fish temperatures at 6 and 24 hours post-harvest. A mean fish temperature profile for all fish monitored at sea was compared with the FDA fish handling guidelines. Temperature profiles for each pelagic fish species were also prepared to estimate the chilling rates.

Data were statistically analyzed using analysis of variance methods (ANOVA, SAS User's Guide, 1985) followed by Least Squares Means analysis to compare specific variable means. Correlation analysis was conducted to analyze the relationship between fish temperature and sea surface water temperature.

Methods Obj. 4. Verify the relationship between post-harvest fish handling procedures and histamine accumulation.

The efficacy of the fish handling methods observed on Hawaii-based commercial troll, handline and longline fishing vessels in Objective 3, for controlling histamine accumulation was evaluated. The mean, range and standard deviation for histamine concentration were calculated for all pelagic fish species combined and for each species group. This comparison determined whether the on-board handling procedures met the FDA handling guidelines, the actual time and temperature parameters achieved and the resulting histamine concentration of fish sampled.

Muscle samples collected from fish with known on-board temperature profiles were analyzed for histamine concentration. The Food Quality Lab (FQL), Honolulu, Hawaii conducted the histamine analyses using the fluorometric method (AOAC, 1995 Official Method 977.13 for Histamine in Seafood). FQL maintained quality assurance and quality control (QA/QC) procedures where single samples were spiked with 1.0 ml of stock histamine solution (1.0 mg/ml) to estimate percent recovery of histamine for each batch of 10 histamine samples tested.

Data were statistically analyzed using analysis of variance methods (ANOVA, SAS User's Guide, 1985) followed by Least Squares Means analysis to compare specific variable means.

Methods Obj. 5. Determine the importance of fish quality grades and odors of decomposition as indicators of histamine concentration.

Additional muscle samples were collected from commercial fish landings at the Honolulu Fish Auction (United Fishing Agency) and the Hilo Fish Auction (Suisan Company Ltd.). These were combined with the fish sampled during the on-board studies (Objectives 3 and 4) to comprise the representative market sample. These two auctions receive and market the majority (an estimated 75 - 90%) of the commercial landings of pelagic fish in Hawaii. Data collected on these fish included fishing gear type, fishing vessel name (kept confidential), date of landing, fish species, weight and quality grade (sensory evaluation). Industry grading methods were used to assign quality Grades No. 1 - 5 to fish, where Grade No. 1 is the highest quality and No. 5 is the lowest quality score. Grades 1 - 4 are acceptable quality fish and the Grade No. 5 category are fish rejected for decomposition. Sensory evaluation for detecting odors of decomposition in the gills and muscle was used to determine if fish should be rejected. Muscle samples were collected, handled and analyzed as previously described.

Data were statistically analyzed using analysis of variance methods (ANOVA, SAS User's Guide, 1985) followed by Least Squares Means analysis to compare specific variable means.

Methods Obj. 6. Develop a HACCP-based strategy for the control of histamine for the fresh tuna industry.

A hazard analysis for histamine poisoning in the fresh tuna industry in Hawaii was prepared by integrating the findings of the Objectives 1 – 5. A Vessel Standard Operating Procedure (VSOP) was drafted for use by the Hawaii fresh tuna industry for the control of histamine.

Methods Obj. 7. Communication of results to the FDA Office of Seafood.

During the course of the project, the PI communicated with staff at the FDA Office of Seafood about the objectives and methods being applied during the project. Dr. George Hoskin, Mr. Don Kraemer, Mr. Walter Staruskewicz and Mr. Jim Barnett were consulted to discuss the project objectives and the research methodologies. FDA inspectors from the San Francisco Office, Ms. Patricia Ziobro, Ms. Darla Bracy and Ms. Jennifer King were also made aware of the project during HACCP inspections of the Honolulu Fish Auction. The implications of the project and its impacts on HACCP controls of histamine in the fresh tuna industry in Hawaii were discussed in detail. The FDA is anticipating the final report in order to make a determination of the validity of the alternative VSOP approach in place at Hawaii's 2 fish auctions for controlling histamine being evaluated by this project.

A workshop directed towards the fishing and seafood industry in Hawaii was held to disseminate the project findings and to get industry feedback for the final report. Another workshop was held for interested personnel from NMFS, the Hawaii Department of Health, and the University of Hawaii involved in seafood safety programs.

B. Project Management: List of individuals and/or organizations actually performing the work and how it was done.

Principal Investigator:	John Kaneko MS, DVM, PacMar, Inc., Honolulu, Hawaii. Designed and managed the project. Conducted some of the vessel-based research. Conducted sensory evaluations, quality grading and collection of muscle samples for histamine analysis. Coordinated statistical analysis. Liaison with SK Program Manager, FDA Office of Seafood staff and seafood industry members. Wrote the final report.
Financial Manager:	Thanh Lo Sananikone, PacMar, Inc., Honolulu, Hawaii. Managed the financial aspects of the project.
Research Assistant:	Donald Hawn, PacMar, Inc., Honolulu, Hawaii. Conducted vessel-based research and assisted in the collection of market samples. Maintained database and assisted in statistical analysis.
Laboratory Services:	Wendy Minor, Food Quality Lab, Honolulu, Hawaii. Conducted the histamine analyses and maintained the laboratory QA/QC procedures.
Statistical Advisor:	Wayne Toma, Statistician, Honolulu, Hawaii. Advised the PI on research design and conducted the statistical analysis.

VI. Findings:

A. Actual accomplishments and findings (corresponding to 7 Objectives).

Results Obj. 1. Evaluate epidemiological data on histamine poisoning in Hawaii.

The Epidemiology Branch of the State of Hawaii Department of Health, provided information on the reported outbreaks of histamine poisoning in Hawaii during the ten-year period between September 20, 1989 and September 28, 1999. Data on reported outbreaks of histamine poisoning in Hawaii were compared with data from the Centers for Disease Control and Prevention (CDC). Discrepancies existed between Hawaii data and the CDC data, which were under reported. Under the advice of Mr. Mits Sugi, epidemiologist with the Hawaii Department of Health, the CDC data was not used in the following analysis.

Table 3. Epidemiology of Histamine Poisoning in Hawaii from September 20, 1989 through September 28, 1999.

Species of Fish involved in outbreaks of Histamine Poisoning in Hawaii (includes confirmed, probable and suspected reports).	No. of outbreaks (%)	No. of illnesses (%)
Common name (Hawaii market name and Latin name)		
Tuna (yellowfin and bigeye) (ahi) (<i>Thunnus albacares</i> or <i>T. obesus</i>)	83 (44.15%)	117 (25.49%)
Mahimahi (mahimahi) (<i>Coryphaena hippurus</i>)	46 (24.47%)	249 (54.24%)
Marlin (Pacific blue and striped) (kajiki and nairagi) (<i>Makaira nigricans</i> or <i>Tetrapterus audax</i>)	15 (7.98%)	24 (5.23%)
Bigeye Scad (akule) (<i>Trachiurops crumenophthalmus</i>)	11 (5.85%)	16 (3.48%)
Wahoo (ono) (<i>Acanthocybium solandri</i>)	7 (3.72%)	12 (2.61%)
Albacore tuna (tombo ahi) (<i>Thunnus alalunga</i>)	1 (0.53%)	2 (0.44%)
Skipjack (aku) (<i>Katsuwonus pelamis</i>)	2 (1.06%)	3 (0.65%)
Canned Tuna (light meat) (<i>K. pelamis</i> and/or <i>T. albacares</i>)	1 (0.53%)	1 (0.22%)
Spearfish (hebi) (<i>Tetrapterus angustirostris</i>)	1 (0.53%)	2 (0.44%)
Mackerel scad (opelu) (<i>Decapterus pinnulatus</i>)	1 (0.53%)	1 (0.22%)
Jack (ulua) (<i>Caranx spp.</i>)	1 (0.53%)	1 (0.22%)
"Covina" (unknown spp.)	1 (0.53%)	1 (0.22%)
Bonfish (oio) (<i>Albula vulpes</i>)	1 (0.53%)	1 (0.22%)
Unknown fish	17 (9.04%)	29 (6.32%)
Total number of outbreaks	188	
Total number of illnesses		459

The reported outbreaks and the number of illnesses due to histamine poisoning in Hawaii are summarized in Table 3. The table includes cases confirmed by histamine analysis of the implicated fish, probable cases based on history and clinical signs and suspected cases which have a lower degree of certainty.

Tuna and mahimahi combined were the most important causes of histamine poisoning in Hawaii during this period causing 68% (129) of the outbreaks and 80% (366) of the total illnesses. The fish species involved in 9% (17) of the outbreaks and 6% (29) of the illnesses could not be identified and are listed in Table 3 as "unknown fish".

The tuna category includes both yellowfin and bigeye tuna because consumers and investigators are often unable to make the distinction. Tuna caused 83 outbreaks representing the highest percentage (44%) of histamine outbreaks in Hawaii. The number of illnesses caused by tuna was only 117 or 25% of the histamine illnesses during this period. Mahimahi was the other dominant species causing 24% (46) of the outbreaks, but a disproportionate 54% (249) of the total number of illnesses.

The origin of the fish implicated in cases of histamine poisoning is extremely important in evaluating histamine risk due to seafood consumption. Twenty-six (26) or close to 14% of the outbreaks of histamine poisoning in Hawaii were found to be caused by imported seafood (Table 4). These outbreaks however, caused a disproportionate percentage (48%) of illnesses (220) indicating a difference between domestic and imported fish in terms of histamine risk. Records indicate that imported mahimahi alone, caused 10% (20) of the outbreaks and an alarming 45% (210) of the illnesses in Hawaii.

Table 4. Histamine poisoning in Hawaii between 9/20/89 and 9/28/99 caused by imported fish and imported mahimahi.

Type of fish	No. of outbreaks	% of total	No. of illnesses	% of total
Imported fish	26	13.82%	220	47.93%
Imported mahimahi (<i>Coryphaena hippurus</i>)	20	10.64%	210	45.75%

The National Academy of Sciences comprehensive study (NAS, 1991) on seafood safety in the US estimated that the highest-risk fish commercially available in the US were imported fresh and frozen fish from tropical areas. The NAS also reported that imported mahimahi was the cause of 47% (55) of the histamine outbreaks in the US between 1978 and 1986. This led the NAS to conclude that embargoing this single product from the US could have a dramatic effect on public health.

The available epidemiological data reflects only those cases that are reported to and investigated by the State of Hawaii Department of Health. From a public health standpoint there are still several important unknowns. The public health impact of fish caught by recreational and subsistence fishers remains uncertain, as these cases of histamine poisoning are likely to be under reported. The NAS (1991) also concluded that recreationally caught fish are likely to be of higher risk for histamine than fish from commercial channels because of inadequate chilling capabilities and lack of awareness of the problem. While government efforts are underway to control histamine in commercial channels through regulatory action, fish from non-commercial sources

remain outside of the reach of the FDA HACCP program. It would be of great interest to evaluate the relative risk of consuming fish caught by commercial versus non-commercial fishers. Epidemiological reports would be of greater value if the distinction between commercial and non-commercial sources of fish was regularly reported. Efforts to control histamine poisoning from non-commercial fish will rely on effective consumer education programs.

Results Obj. 2. Develop Fleet on-board handling profiles (trollers, handliners and longliners).

Fleet profiles were prepared briefly describing the gear, fishing methods, the typical on-board fish handling procedures, the fish species caught and quality factors.

Trolling fleet profile.

Fishing method (trolling).

Trolling entails the use of fishing rods or handlines rigged with artificial lures or natural baits, which are towed near the surface behind a moving boat. Trolling is an "active" fishing method where the fish is enticed to strike a lure or bait in motion simulating a live prey fish. In Hawaii, trollers typically troll with 4 to 6 lines from the stern and from outrigger poles, which extend laterally from the vessel.

Trolling gear in Hawaii is used during daylight hours and most trollers begin the day at sunup and return to the dock by sundown. Hawaii trollers described here are engaged in single-day trips and deliver fresh fish stored in ice.

Project researchers went on 7 commercial trolling trips to document the fishing and fish handling methods as well as collect data needed for other project objectives. When a fish is hooked, the line (and fish) is retrieved quickly in order to minimize the length of time the fish struggles. The time on the line is generally less than 5 minutes, however this depends on the efficiency of the fisher. If the troller is operating as a charter vessel, the angler may take considerably more time (60+ minutes) to retrieve fish especially when large fish such as marlin are caught. In the State of Hawaii, fish caught by charter boats may be sold under the commercial fishing license of the vessel operator.

On-board fish handling methods (trolling).

Once the fish is brought to the side of the boat it is gaffed and stunned with a club. The fish is then brought on-board where it is immediately bled with knife cuts to the gill arches, under the pectorals or at the caudal peduncle near the tail. The fish is bled for 5 to 10 minutes while being rinsed with clean seawater and then placed into an insulated fish box containing an ice slurry made up of approximately 2 to 3 parts ice to 1 part seawater. Generally, fish are left whole and are not dressed at sea. The steps from stunning the fish to being placed in the fish hold take no more than 15 minutes.

Great attention is placed on monitoring the condition of the ice slurry during the trip. Too little ice and the fish may not cool rapidly and the fish skin will be abraded with the constant movement of the boat. The outward appearance of fresh fish in Hawaii is very important to the perception of quality by the buyers and has a significant effect on the

eventual sale price (Bartram, et al, 1996). Fishermen constantly adjust the ice and seawater mixture in order to maximize the cooling rates and the quality of the fish. Special care is given to individual fish because they are priced and sold based on individual quality mainly through Hawaii's 2 display auctions.

By the time the vessel returns to the dock, the first fish caught in the morning may have been in the ice slurry (and later in ice alone) for over 10 hours. However, the last fish caught may have only been in the ice slurry for 1 hour. This accounts for the range of fish temperatures observed at the time of landing by Hawaii's troll fleet. Fish are then kept in ice, transported to the auctions, weighed, their temperature recorded and then placed in the auction cold storage rooms and buried in ice overnight for sale the following day after the fish are finally chilled to below 40° F. Alternatively, fish that are less than 24 hours out of the water are kept in ice and sold at the auction with the buyer accepting the responsibility to continue to properly chill the fish.

Fish caught and quality factors (trolling).

Hawaii's troll fleet catches mahimahi, marlins, tuna, skipjack and wahoo (Boggs and Ito, 1993). Trollers fish single-day trips and produce fish that are only 1 day out of the water. This fleet produces the highest quality fresh mahimahi, marlins and wahoo available to the Hawaii fresh fish market. These troll-caught fish routinely receive premium prices over fish caught by the other gear types and especially over imported fish. This is in sharp contrast to troll-caught yellowfin tuna which are not considered to be high in quality or long in shelf-life, presumably due to the capture methods, which involve a struggle on the line before death. Troll-caught tuna are susceptible to rapid muscle color change from red to brown resulting in a greatly reduced market value.

During the summer months troll-caught yellowfin tuna may also tend to be affected by the "burnt tuna syndrome" (Nakamura, et al., 1987). The affected muscle turns pale, opaque, watery and soft in texture making the fish unsuitable for sashimi and other higher quality, higher-value preparations. Subsequently the value of burnt tuna is reduced considerably.

Both quality problems (rapid color change and burnt tuna syndrome) associated with troll-caught tuna may be related to the fishing method which involves a struggle on the line, elevated body temperature and other physiological factors at the time of death and temperature controls in the post-harvest handling period. There are also likely to be multiple pre-disposing factors that trigger the burnt tuna defect.

Handline fleet profile.

Fishing method (handline).

The typical handline used in Hawaii to catch tuna consists of a nylon rope connected to a mainline of polypropylene or Dacron, which is attached to a leader of monofilament nylon ending with a single baited hook. Each boat deploys 4 handlines and drifts in the current in areas known to aggregate tuna. Parachute sea anchors are used to control the drift.

The Hawaii handline fleet is comprised of two segments. The first operates nearshore and mostly at night during the summer run of yellowfin tuna. These handline fishers

leave the dock in the late afternoon and once on the fishing grounds, set the sea anchor and begin a drifting pattern. Fishing continues from sundown until sunup. Once a fish is hooked, the line is retrieved by hand (no reels involved). This takes less than 10 minutes with some fishers able to retrieve, bleed, gill and gut and place fish into an ice slurry in less than 10 minutes from the time of hook-up (Nakamura et al, 1987). After returning to the harbor, the fish are unloaded and delivered to the fish auctions or directly to wholesalers.

The second segment of the Hawaii handline fleet focuses on fishing at offshore seamounts and weather buoys which tend to aggregate small to medium-sized bigeye and yellowfin tuna. The offshore handline fishing fleet differs slightly in that the trips are longer (2-5 days) and the fish are caught at the surface with the handlines. This method is similar to the pole and line or bait boats that harvest actively feeding schools of tuna at the sea surface.

On-board fish handling methods (handline).

Once the fish is brought to the side of the boat it is gaffed and then stunned using a club or a pistol. The fish is then brought on-board with gaffs and bled using knife cuts at the gill arches, under the pectorals or at the caudal peduncle. After a few minutes (5 – 10 minutes) of bleeding while being rinsed with clean seawater, the fish may be gilled and gutted, headed and gutted or left whole and placed into insulated fish boxes containing an ice/seawater slurry.

Attention to the fish in the ice slurry is essentially the same as with the trollers. For the nearshore handline fishery, the first fish caught in the evening may have been in the ice cooling for 12 hours, while the last fish caught might have had less than 2 hours to chill by the time they are landed. This accounts for the range of delivery temperatures for handline fish. In Hilo, Hawaii, the center of the summer nearshore yellowfin handline fishery, these tuna are often sold at auction immediately after landing. Fish are kept in ice and the auction and buyers (wholesalers) assume the responsibility for continuing to properly chill the fish after delivery from the vessel. For handline fishing at the offshore seamounts, many of the fish are over 24 hours on ice by the time they are delivered and should therefore be below 40° F.

Fish caught and quality factors (handline).

The nearshore handline fleet catches primarily yellowfin, bigeye, albacore and an occasional swordfish (Yuen, 1979). Handliners fishing nearshore generally fish short trips and deliver fish to the market that are from a few hours to 2 days out of the water. The quality of handline-caught tuna is generally intermediate between that of troll-caught tuna (lower quality) and longline-caught tuna (higher quality possible). Although the quality of handline tuna can be very good, the total shelf life of these fish is greatly reduced when compared with longline-caught fish. The muscle color of handline yellowfin tuna tends to change rapidly from red to brown. Both handline and troll-caught tuna are also prone to the effects of the burnt tuna syndrome during the summer months (Nakamura et al, 1987).

The offshore handline fishery tends to catch more small to medium-sized bigeye tuna and some yellowfin in contrast to the nearshore fishery that catches primarily large yellowfin. Offshore handliners deliver fish that are mostly on ice for over 24 hours. The

bigeye caught by this fleet are generally lower in quality, have a shortened shelf life and tend to have soft muscle texture.

Longline fleet profile.

Fishing method (longline).

Longlining entails the use of a long monofilament longline (5 to 40 miles long) with multiple leaders and baited hooks (200 to 1500 hooks). The average number of hooks fished per day for Hawaii's longline fleet (both tuna and swordfish trips) in 1998 was 1,390 (Ito and Machado, 1999). Each time the longline is deployed and retrieved is called a "set". Although each fishing vessel may adopt different strategies for when and where to fish, the set begins with deploying one end of the longline that is attached to a float fixed with a flag and radio beacon. The set is completed when the last hook remaining in the water is retrieved. Fishing trips targeting tuna typically range from 14 to 21 days, while trips targeting swordfish range from 30 to 45 days (WPRFMC, 1995).

As the vessel moves forward, the longline is deployed off the stern and the crew attaches leaders and baited hooks at intervals between additional floats and flags used to mark the location of the line. Deploying the line typically takes 4 hours but may take up to 6 hours depending on the amount of hooks fished per set. After the line is deployed, the crew may take a break to "soak" the line before starting the retrieval.

The mainline is deployed so that the hooks are at a depth ranging from 50 to 400 meters depending on targeted species, the position of the hook along the mainline and the skill of the crew and captain. Once a fish is caught it may remain alive on the line until retrieval. Other fish may struggle against the line and expire by the time they are brought to the vessel. Retrieving the line can take an average of 8 hours depending on the number of fish caught and the sea and weather conditions.

On-board fish handling methods (longline).

Once brought to the vessel, the fish are gaffed and hauled on deck. Live fish are stunned, brain spiked (pithed) and then bled. Fish that are retrieved dead are also bled using cuts to the gill arches, under the pectoral fins and/or at the caudal peduncle. After 5 -10 minutes of bleeding while the fish is rinsed with clean seawater, the fish may be gilled and gutted or left whole. The fish are then taken below deck into the fish hold and buried in ice. Longline fishers take great care in handling fish to ensure optimum outward appearance, muscle quality and marketability. Unlike the handling typical of trollers and handliners, Hawaii longliners for the most part do not use ice slurries (ice brine) to pre-chill the fish before placing them in ice.

As the fish cools, the surrounding ice melts. The space that forms creates an insulating layer of air or "igloo effect" that greatly reduces the heat transfer efficiency. Direct contact with the ice is needed to maintain optimum cooling rates. Repacking fish in the ice takes place anywhere from 3 hours to 24 hours after first being placed in the fish hold. Fish are stored buried in ice for the remainder of the trip. Only a few of the longline vessels in Hawaii have icemakers and/or refrigerated fish holds. Most of the fleet uses only the ice they have at the start of the trip and depend on insulated fish holds to keep the ice from quickly melting.

Fish caught and quality factors (longline).

Hawaii's longline fleet catches bigeye tuna, yellowfin tuna and swordfish as the primary target fish species. In addition, the fleet catches multiple species of commercially important pelagic fish including albacore tuna, skipjack, Pacific blue and striped marlins, mahimahi, wahoo, spearfish, moonfish, pomfrets and sharks (Boggs and Ito, 1993). Longline-caught fish vary in the length of time out of the water and in quality because of the fishing method, the number of sets and length of the trips. Some of the longline caught bigeye tuna are extremely high quality and value (over \$17.00/lb round weight basis occurs) while other fish of the same species from the same vessel trip may be rejected from sales at the time of unloading due to decomposition. The entire range of fish quality is possible within the same load of fresh fish caught by longliners and can be quite varied within sets, between trips and between vessels. Random representative sampling is not used by buyers to judge the quality and value of the load because it is not effective in predicting the quality of the individual fish in the catch.

In other locations in the US longline-caught fish are sold on a "boat run" basis with an average price negotiated for the entire load or by broad quality grade categories. By contrast, the fresh tuna industry in Hawaii sells fish on an individual basis and relies on judging the quality of each fish. Every fish is screened for quality attributes (muscle color, clarity, texture and fat content) and as quality grade declines, buyers look more closely for signs of decomposition.

The display auction system in Hawaii allows the buyers to closely inspect individual fish before bidding. Fish quality is a primary determinant of price in the fresh tuna market in Hawaii (Bartram, et al. 1996). Prices are determined by competitive open bidding and prices rise by \$0.10/lb increments. The 2 auction houses in Hawaii take responsibility to receive, screen and cull fish for signs of mishandling and decomposition. Once the fish are displayed, the buyers must again carefully screen the fish for signs of mishandling, quality defects and indications of decomposition in order to decide on market value. Buyers are keenly aware of subtle quality differences that result in the range of prices paid. Auction prices for fish can range from over \$17.00/lb down to \$0.10/lb round weight basis, depending on the quality, species of individual fish and market conditions.

Results Obj. 3. Verify the post-harvest fish handling procedures.

In order to verify the post-harvest fish handling procedures, the standard operating procedures on fishing vessels representative of commercial longline, troll and handline fishing practices were monitored during research trips on commercial fishing vessels during normal operations. On-board research consisted of 7 single-day trips on commercial trolling vessels, 5 single-day (overnight) trips on handline vessels and 21 longline sets during 14-day and 22-day longline trips made by a single vessel.

The results are presented in order to answer key questions about handling procedures.

How much time are fish dead on the line?

Trollers.

None. Troll-caught fish are brought to the boat alive. Although troll-caught fish may not die on the line, it is of interest to know how long the fish might struggle because of the

physiological effects on body temperature, energy stores, the onset, strength and duration of *rigor mortis* and the postmortem degradation processes. The time observed fighting on the line averaged 6 minutes with a minimum of 1 minute and a maximum of 30 minutes. The time it takes to get the fish on the boat after hooking depends on the species of fish, its size, the strength of the fish and fisher and the efficiency of the gear. Time on the line is expected to vary greatly on charter boats where anglers are non-professional and may not be able to or interested in quickly retrieving the fish.

Handliners.

None. Handline-caught fish are quickly brought to the boat alive. Fish caught on handlines struggle and the time observed on the line was an average of 4 minutes with a minimum of 1 minute and a maximum of 10 minutes. This is within the range of handling parameters reported by Nakamura et al (1987) in the Hawaii handline fleet. The time fish are on the line depends on the species of fish, the size of the fish and the efficiency of the fisher. Handline fishers are generally aware of the need to minimize the time the fish struggles to maintain fish quality.

Longliners.

None for live fish. For dead fish, up to 20 hours is possible. The time and temperature data reported for fish (dead and alive) monitored during this study began at the time the temperature loggers were placed in the muscle after being brought on-board. For a fish that is retrieved alive, "time zero" is when the fish is brought on-board. For fish that die on the line during the set, time zero occurs some time after the hooks are deployed and when the fish dies before being retrieved.

Because of the way in which longline gear is typically deployed and retrieved, there are concerns about the total length of time that fish might be hooked and remain on the line until being retrieved. The time on the line is important because of the potential for histamine formation in fish that die in warm tropical and subtropical waters.

It is not possible to determine the exact time of death on the line without sophisticated equipment. The discussion of chilling rates in later sections of this report does not include the additional time on the line for fish retrieved dead. There have been attempts to estimate the time of death by evaluating the fish temperature at different locations in the body at the time of retrieval, but this method proved inadequate without accurate water temperature and depth of capture data (Pages, 1972). Longline research in Hawaii using hook timers and Time/Depth Recorders (TDRs) attached to longline hooks, determined that bigeye tuna and yellowfin tuna survive much longer after being hooked than previously thought (Boggs, 1992). Over half of the bigeye survived 9 hours after being hooked and the shortest time recorded for death after hooking was 2 hours.

The maximum possible time for a fish to be dead on the line is from the time the first hook enters the water at the start of the line deployment to the time the last fish is retrieved. For the 21 longline sets monitored during the study, the mean maximum possible time was 18 hours (Table 5).

The shortest amount of time possible for fish to be dead on the line is in situations when the fish dies immediately before being hauled on-board. Another more practical determination is the "soak time" for the longline gear. This is the lapsed time between

the end of the deployment of the longline gear and the beginning of the line-hauling phase.

Table 5. Soak time and time dead on the line.

No. of sets	Soak time for longline gear. (hours)				Maximum amount of time dead on the line. (hours)			
	mean	SD	Min	max	mean	SD	min	max
21	6.74	0.74	5.67	8.15	18.02	1.23	16.12	20.48

How many fish came up alive and how many were dead?

Trollers.

All fish were alive. Trolling gear entices the fish to strike live or dead bait, or artificial lures and all fish are hooked and brought to the boat alive.

Handliners.

All fish were alive. Handline fishers use baited hooks and all fish are hooked and brought to the boat alive.

Longliners.

Some were alive and some were dead. Table 6 displays the breakdown of longline-caught fish observed during the study retrieved alive or dead. Fish from 21 longline sets were evaluated. Bigeye tuna, yellowfin tuna, albacore, striped marlin, blue marlin and mahimahi were sampled from the longline catch because these are the primary market species susceptible to forming histamine. A total of 383 pelagic fish were observed. Of that total, 152 (39.7%) were alive and 231 (60.3%) were dead when brought on-board.

Longlines are set, soaked and retrieved over an extended period of time from 16 to 18 hours from start to finish. What determines if a fish will be alive or dead, depends on the species of fish, the amount of time it stayed on the line and whether it struggled against the line (and died) or was calm and survived until the line hauling began.

What was the deck time for dead and alive fish?

"Deck time" is of interest in estimating the time it takes fishers to pre-process each fish from the time it is brought on-board until it is placed in ice in the fish hold. The fish are vulnerable at this time to elevated ambient temperature on deck and to bacterial contamination. Deck time should be kept to a minimum necessary to properly pre-process the fish prior to chilling and storage. Efforts to maintain proper sanitation and prevent contamination are also extremely important.

Trollers.

The average deck time observed was 7.2 minutes (range 3.0 to 10.0 minutes).

Handliners.

The average deck time observed was 8.6 minutes (range 3.0 to 20.1 minutes).

Longliners.

The average deck time observed for live versus dead fish was observed and recorded for longline fish. Table 6 displays the total amount of time it took the longline crew to place fish into the ice after being brought aboard. The average deck time for all species (alive and dead) was 12.2 minutes, with a minimum of 0 minutes and maximum of 83 minutes (SD = 11.4 minutes).

The time it takes for fishers to bleed and process fish prior to placement in the ice depends on many factors including the catch rate, any delays due to mechanical complications with retrieving the gear, weather and sea conditions, the species and size of fish, etc. The average deck time for dead fish across species was 12.0 minutes with a minimum of 0 minutes and a maximum of 83 minutes (SD = 12.4 minutes). The average time for live fish across species was 12.6 minutes, with a minimum of 1.0 minute and a maximum of 63.0 minutes (SD = 9.5 minutes).

Table 6. Deck time for live and dead tuna, marlin and mahimahi caught by longline.

Common name		N	%	Deck time (minutes)			
				mean	minimum	maximum	SD
Bigeye tuna	all	86		11.9	0.0	28.0	6.5
	live	41	47.7	13.4	4.0	27.0	6.9
	dead	45	52.3	10.6	0.0	28.0	5.8
Yellowfin tuna	all	29		12.0	0.0	29.0	7.8
	live	10	34.5	15.3	7.0	22.0	4.8
	dead	19	65.5	10.2	0.0	29.0	8.5
Albacore tuna	all	103		7.7	1.0	57.0	6.9
	live	26	25.2	7.9	2.0	24.0	4.7
	dead	77	74.8	7.7	1.0	57.0	7.5
Striped marlin	all	73		16.6	1.0	83.0	15.0
	live	17	23.3	16.7	8.0	34.0	7.9
	dead	56	76.7	16.6	1.0	83.0	16.6
Blue marlin	all	5		26.2	10.0	54.0	15.3
	live	2	40.0	18.5	17.0	20.0	1.5
	dead	3	60.0	31.3	10.0	54.0	18.0
Mahimahi	all	87		13.5	1.0	63.0	14.0
	live	56	64.4	12.3	1.0	63.0	12.6
	dead	31	35.6	15.6	1.0	62.0	16.0
All species	all	383		12.2	0.0	83.0	11.4
	live	152	39.7	12.6	1.0	63.0	9.5
	dead	231	60.3	12.0	0.0	83.0	12.4

Fishers are generally aware of the need to quickly prepare the fish, minimize the deck time and begin the chilling process. It is possible that fishers may prioritize the handling of pelagic fish based on the value of the fish species and possibly the susceptibility of the particular species to quality defects related to handling and temperature controls. The maximum deck time for the high value fish species, bigeye and yellowfin tuna is

relatively low compared with the two marlin species and mahimahi which generally receive significantly lower prices in the market even for high quality fish.

It should be stressed that the observed maximum deck time of 83 minutes was an unusual situation and occurred when a striped marlin was retrieved and the mainline became tangled in the propeller shaft in rough seas, greatly extending the deck time.

What was the initial core body temperature at the time of boarding for live and dead fish caught by longline?

The initial core body temperature of 134 mixed pelagic fish caught by longline gear was measured and recorded in order to establish a baseline for live fish and those that died on the line. The initial body temperature at the time the fish were brought on-board determined the magnitude of the temperature drop required to properly chill the fish. The single blue marlin monitored was not included in this comparison.

The results are presented in Table 7. Fish that died on the line tended to be 10° F colder than those that were retrieved alive. Within each species group, the mean initial core temperature of dead fish was significantly lower than for fish brought up alive. Live fish across all species had a mean initial core temperature of 79.54° F. Live fish struggle as the line is hauled and tend to have an elevated body temperature and a greater temperature drop required for proper chilling.

Table 7. Comparison of initial core temperature of pelagic fish caught by longline retrieved alive and dead.

Fish species	Live fish Initial core temp (° F)			Dead fish Initial core temp (° F)			Probability
	N	mean	SD	N	mean	SD	
Bigeye	17	78.71	1.54	13	65.14	8.16	0.0001
Yellowfin	9	79.81	1.64	10	71.43	6.96	0.0027
Albacore	14	80.25	5.07	34	67.76	7.60	0.0001
Striped marlin	8	79.59	1.25	13	74.49	5.81	0.03
Mahimahi	12	79.64	1.02	4	70.78	8.00	0.0013
All species	60	79.54	2.69	74	69.14	2.69	0.0001

Fish that died on the line had an average core temperature of 69.14° F. Dead fish tended to have a lower body temperature than live fish presumably because of the heat transfer to the water at the relatively cooler temperature found at the hooking depth.

Experimental longline fishing within the area in the central North Pacific typically fished by Hawaii's longline fleet confirmed the highest catch rate for bigeye tuna at 360 to 400 meter depths where water temperatures range from 46-50° F (Boggs, 1992). Bigeye are known to aggregate in water depths where corresponding water temperature is in the range of 46-50° F. This is fortuitous because if the fish dies, it is in water that is cold enough to begin the chilling process immediately. Histamine formation is known to be rapid at temperatures above 70° F and especially high at close to 90° F (FDA, 1998). The relatively cool temperatures at the hooking depth, even in tropical Pacific waters, may be one of the reasons histamine accumulation is not a more common problem with longline-caught tuna and other pelagic species in Hawaii.

The initial core temperature of fish is presumed to be correlated with water temperature. The correlation between sea surface temperature (SST) and the initial core temperature was analyzed for each of the pelagic species monitored during the 21 longline sets. SST was recorded at the beginning of each set and compared with initial core temperature of live and dead pelagic fish. SST is recorded by a thermistor placed on the vessel's hull, 9 to 12 feet below the sea surface and is readily available information on commercial longline vessels. Correlations for all pelagic fish except the 1 blue marlin are reported in Table 8.

Initial body temperature correlated with SST only for bigeye and striped marlin brought on-board alive. For live bigeye tuna the correlation coefficient was 0.62 ($P < 0.01$) and for live striped marlin the correlation coefficient was 0.82 ($P < 0.01$). Correlations may exist between initial core temperature of the other categories of fish and the water temperature at the depths where fish are hooked and not the sea surface. The time the fish is held at the particular water depth (and water temperature), species anatomical and physiological differences and fish size are other potentially important variables.

Table 8. Correlations between sea surface water temperature (SST) and initial core body temperature of pelagic fish brought on-board alive and dead during longline sets.

Fish	Alive		Dead	
	SST vs Initial Core Temp Corr. coef.	Probability	SST vs Initial Core Temp Corr. coef.	Probability
Bigeye	0.62	0.01	0.28	0.35
Yellowfin	0.22	0.57	0.49	0.15
Albacore	0.13	0.66	-0.05	0.77
Striped marlin	0.82	0.01	0.06	0.84
Mahimahi	0.48	0.11	0.83	0.17
All species	0.11	0.36	0.13	0.22

What was the temperature profile for fish stored in ice?

After the initial core temperature of the fish was recorded, the temperature loggers were placed in a sample set of 80 mixed pelagic fish to record detailed time and temperature histories during the remaining period of ice storage on the longline vessels.

The key parameters considered were, the temperature after 6 hours, the temperature after 24 hours, the time to below 50° F and the time to below 40° F. Using the combined data from all of the fish (dead and alive) monitored in longline sets with detailed temperature histories, the average core temperature at 6 hours was 46.15° F (Figure 2). The average core temperature at 24 hours was 33.2° F. Disregarding the additional time for fish that died on the line, on average, these longline-caught fish were handled on-board in compliance with the FDA handling guidelines. Once they were brought aboard fishers were capable of chilling fish to below 50° F within 6 hours and to below 40° F within 24 hours to control histamine accumulation.

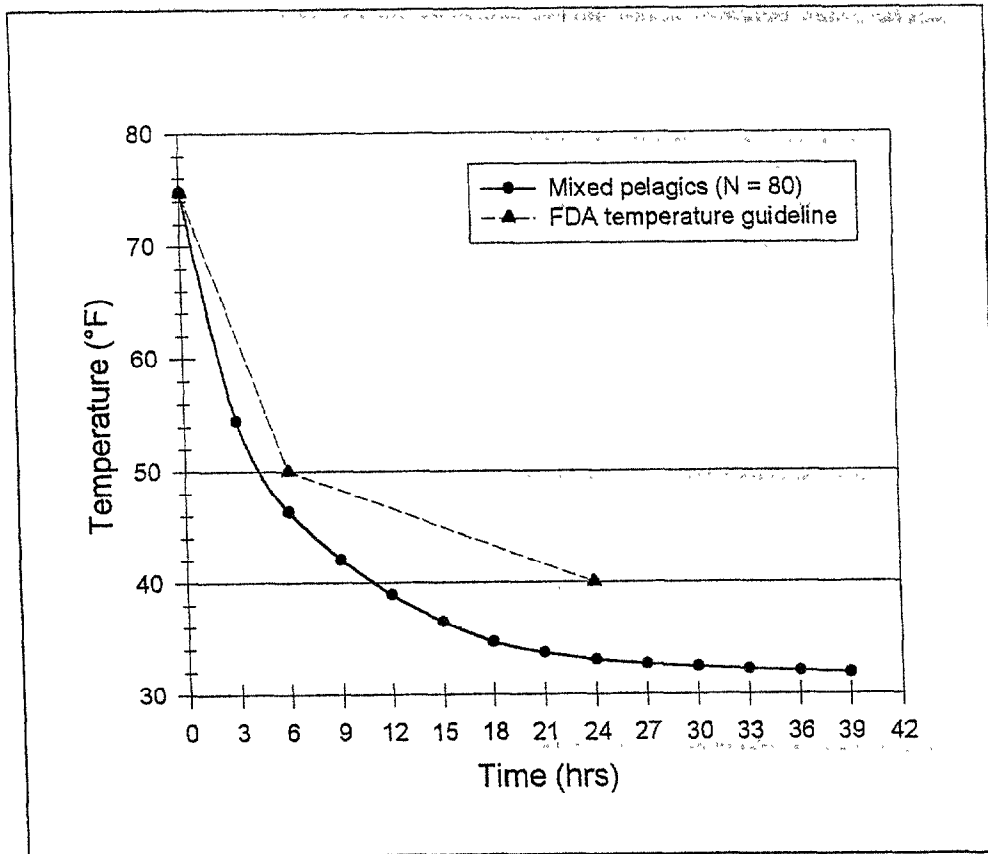


Figure 2. Mean On-board Fish Temperature Profile for mixed pelagic fish retrieved dead and alive by longline gear, chilled and stored in ice.

The chilling patterns for each species were also evaluated. Chilling patterns for bigeye, yellowfin, albacore are presented in Figure 3, and for striped marlin, blue marlin and mahimahi in Figure 4.

Bigeye tuna brought aboard alive, did not meet the 6-hour to below 50° F guideline, but were below 40° F within 24 hours after death. After 6 hours, live bigeye were 51.2 ° F and after 16 hours these fish were below 40° F. Dead bigeye began the on-board chilling sequence at about 65° F, were below 50° F in just over 4 hours and below 40° F within 13 hours after boarding.

Yellowfin tuna brought to the boat alive did not meet the 6 hours to 50° F guideline, but met the 40° F guideline well within the 24-hour period after death. After 6 hours, live yellowfin were a few degrees above 50° F and after 14 hours were below 40° F. Dead yellowfin, did not meet the 6-hour to 50° F guideline, but met the 40° F guideline within 14 hours after being brought on-board. The initial temperature difference between live and dead yellowfin was made up within the first 4 hours of chilling.

Albacore tuna brought to the boat alive easily met both the 40° and 50° F guidelines. Dead albacore were chilled to below 40° F after 2 hours and to below 40° F within 8 hours of boarding.

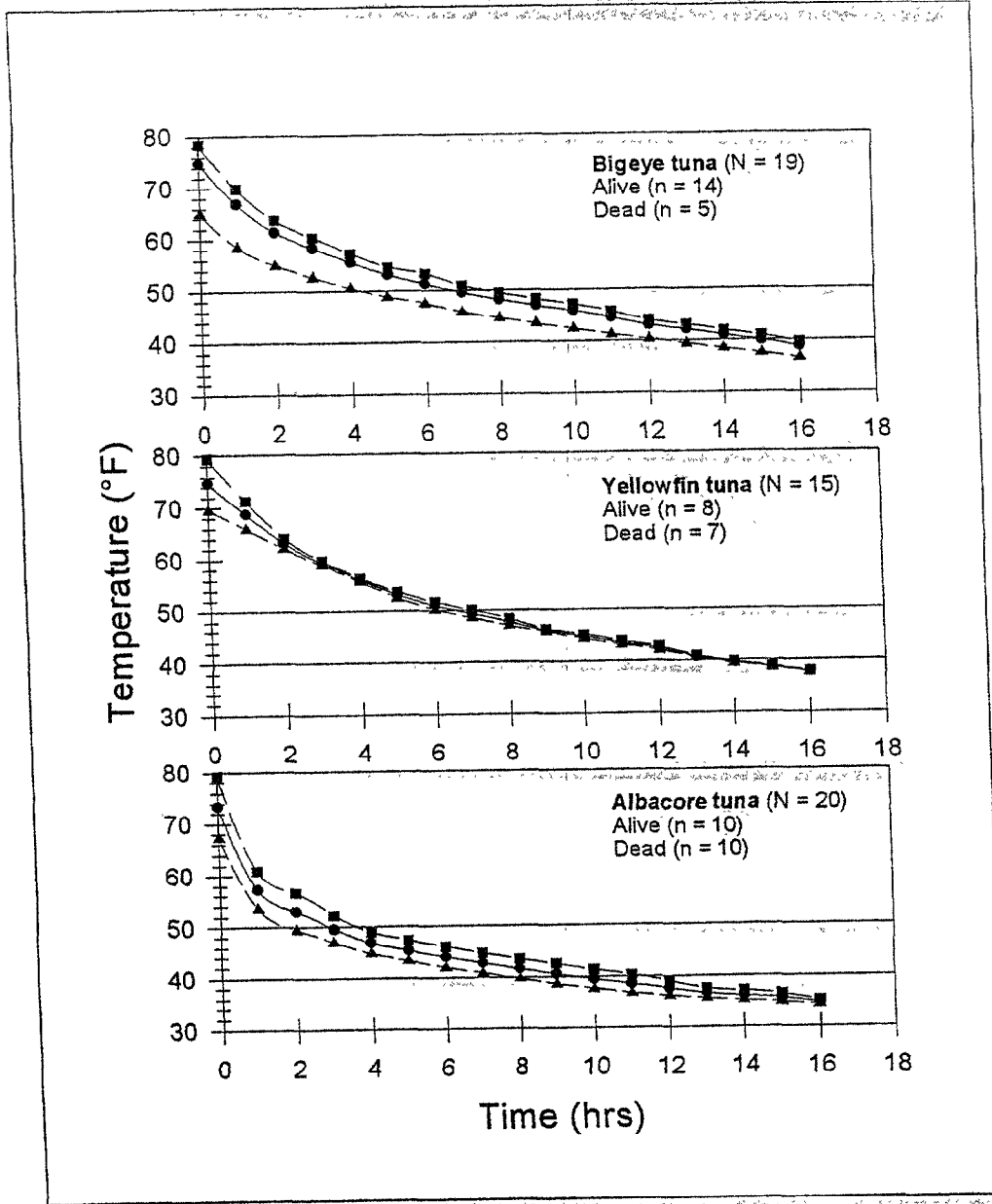


Figure 3. Fish Temperature Profiles (chilling patterns) for bigeye, yellowfin and albacore tuna caught by longline. (alive=■, dead=▲, all=●)

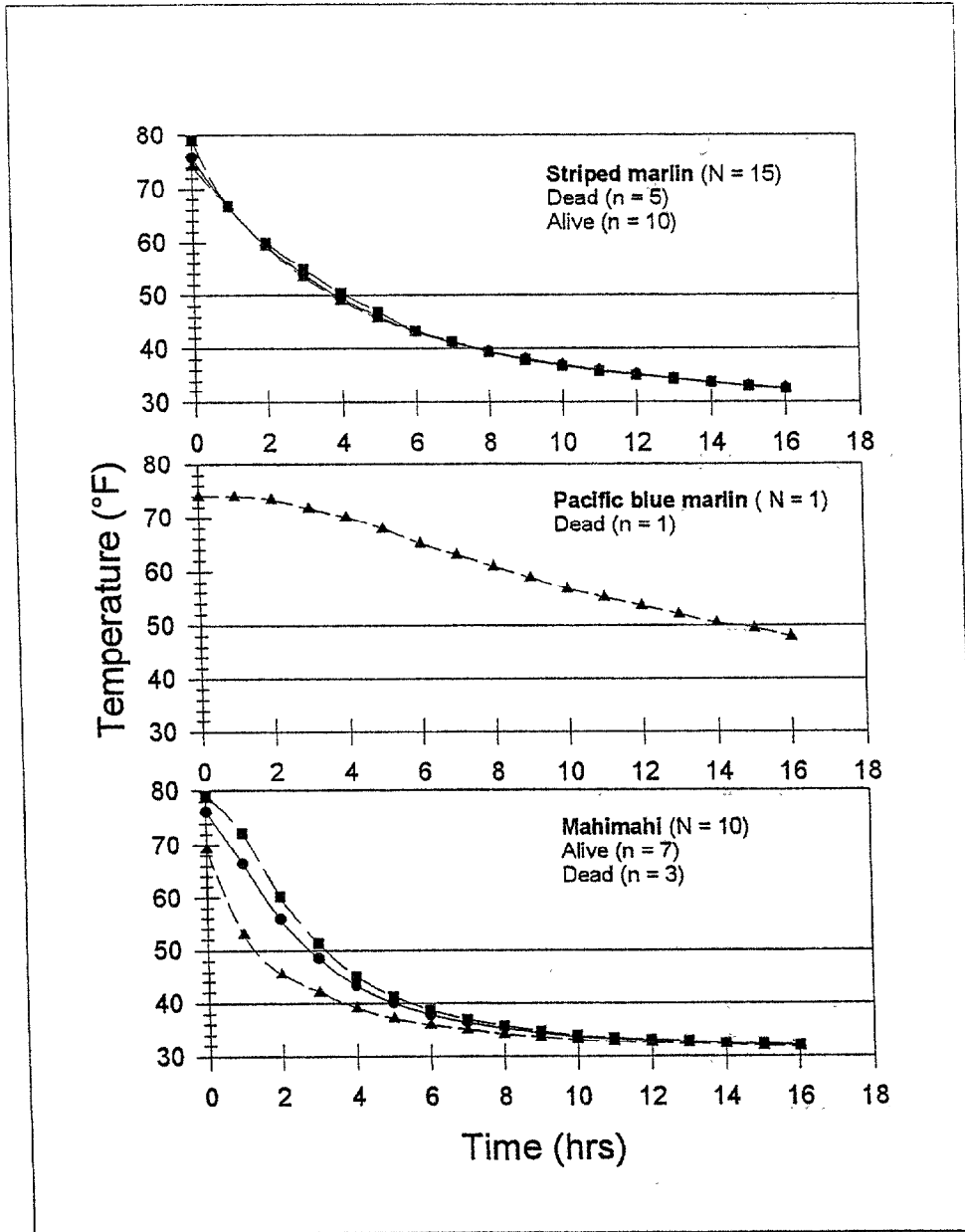


Figure 4. Fish Temperature Profiles (chilling patterns) for striped marlin, Pacific blue marlin and mahimahi caught by longline. (alive=■, dead=▲, all=●).

Striped marlin brought to the boat alive were chilled to below 50° F after 4 hours and to below 40° F after 8 hours after death. Dead striped marlin were chilled to below 50° after 4 hours and to below 40° F within 8 hours.

A single Pacific blue marlin was monitored at sea. This fish died on the line. After boarding, this fish took over 14 hours to be chilled to below 50° F but dropped below 40° F within 24 hours. This fish was large (369 lbs round weight), did not meet the 50° F guideline but was cooled to below 40° F, 23.9 hours after being brought aboard.

Mahimahi brought to the boat alive were chilled to below 50° F after 3 hours and to below 40° F within 6 hours after death. Live mahimahi complied with both of the FDA guidelines. Mahimahi that died on the line were chilled to below 50° F after 1 hour in ice and to below 40° F within 4 hours after boarding.

Did the fish chilling method make a difference?

The chilling method is of potential importance. Hawaii handliners and trollers tend to use an ice slurry to pre-chill fish before storage in ice alone. By contrast, longliners tend to pack fish directly into ice and after an initial cooling period, they repack the fish in ice to be sure that the ice is in full contact with the fish skin and that cooling proceeds effectively.

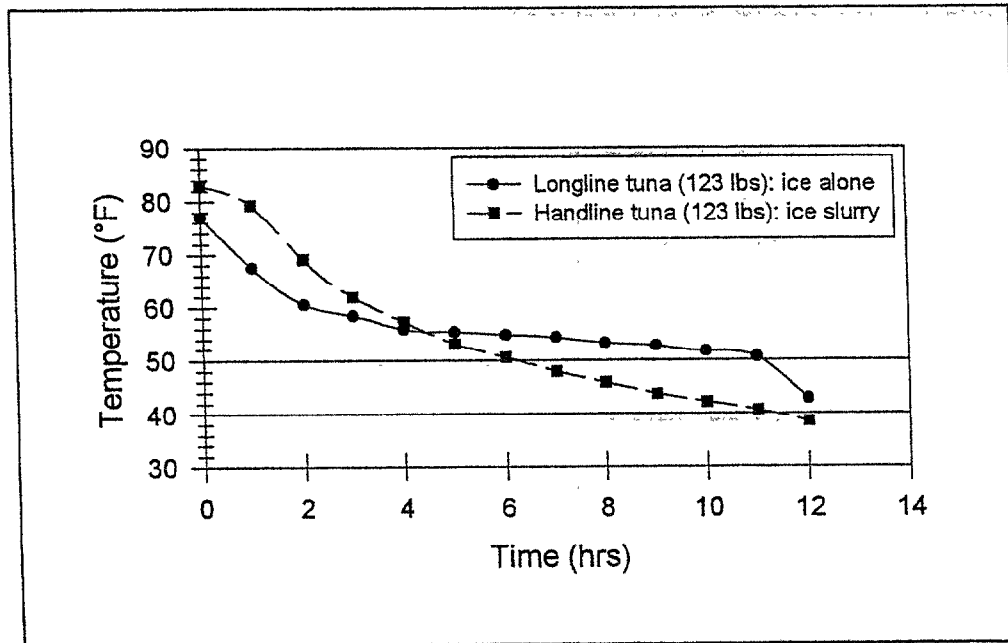


Figure 5. Fish Temperature Profiles (12 hours) comparing chilling rates for individual tuna (123 lbs each) caught by longline and handline, chilled by ice alone and ice slurry.

The comparison of these chilling methods in Figure 5 illustrates the difference in heat transfer efficiencies between ice alone and ice slurries (seawater and ice). Both tuna were of equal weight and were both brought to the vessel alive. The initial body temperature differed with handline-caught fish being higher (82.9° F) than the longline-caught fish (77.0° F). Although the handline fish started the chilling process at a higher temperature, the ice slurry method was much more efficient in heat transfer and after 4 hours, the handline fish was cooler than the longline fish. The chilling rate in the ice slurry continued to be faster, bringing the handline fish to 50° F after 6.2 hours and to below 40° F after 11.1 hours.

The longline fish held in ice alone had a much different cooling pattern. The temperature dropped steeply for the first two hours. After that, the cooling rate slowed greatly with little change until a steep temperature drop occurred after 11 hours. Initially the ice was in direct contact with the fish and chilling was efficient. After two hours, the ice melted immediately adjacent to the fish, forming an air space surrounding the fish. The heat

transfer capacity of cold stagnant air is much less than water (20X) and the heat transfer rate slows substantially. After 11 hours, the crew repacked the fish in ice to ensure direct contact between the fish and the ice and the chilling rate accelerated greatly. By hour 12, the temperature differential between the two fish was almost eliminated. This temperature profile suggests that longline crews may want to carefully reconsider the time they wait before repacking fish in the ice. Repacking after a shorter waiting period would help to maintain optimum heat transfer and overall chilling rates. The advantages of a shorter waiting period should be weighed against the possibility that a second repacking might become necessary. The cooling efficiency of an ice slurry over ice alone is potentially significant in terms of fish quality and food safety because it is an extremely effective method of heat transfer during the critical handling period when fish are $>70^{\circ}\text{F}$.

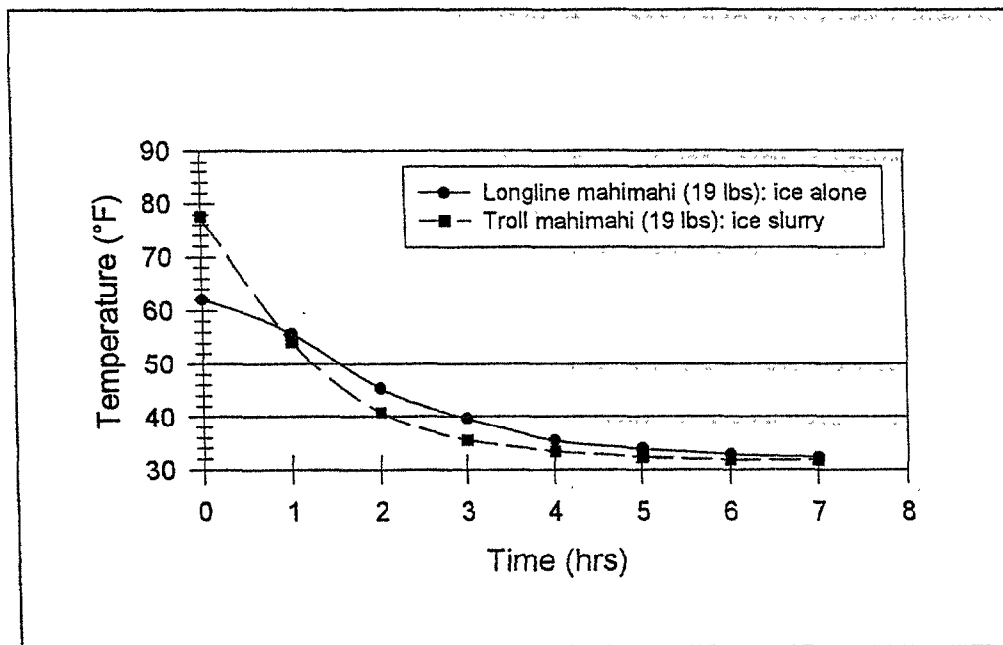


Figure 6. Fish Temperature Profiles comparing chilling rates for individual mahimahi (19 lbs each) caught by longline and troll gear, chilled by ice alone and ice slurry. The troll fish was alive and the longline fish was dead upon retrieval.

The difference between chilling methods on mahimahi of similar size is illustrated in Figure 6. The troll-caught mahimahi was landed alive and chilled in an ice slurry, had an initial core temperature of 79°F and yet after 1 hour, was cooler than the longline mahimahi stored in ice. Note that both mahimahi were chilled at relatively fast rates compared to the tuna in Figure 5. Body size (weight) accounts for much of this difference. However, the body conformation also impacts the heat transfer rates. Mahimahi are compressed laterally and have a greater surface area for heat exchange, while tuna are more rounded in cross-section and are anatomically evolved for heat retention.

What were the vessel standard operating procedures for post-harvest fish handling?

Vessel standard operating procedures for post-harvest fish handling were observed and monitored for fish brought aboard alive and dead. The parameters monitored included

fish weight, initial core temperature, deck time, time to 50° F, time to 40° F, temperature at 6 hours (after boarding), temperature at 24 hours (after boarding), the total fish hold time, and at the end of the trip the quality grade and corresponding histamine concentration. Post-harvest handling data were summarized for each species group and for all fish combined (Table 9a and 9b). The means of each parameter were compared across fish species groups.

Table 9a. Verification of post-harvest fish handling during longline fishing sets: fish weight, initial core temperature, deck time, time to 50° F and time to 40° F.

FISH	WEIGHT (lb.) (p < 0.0001)			INITIAL CORE TEMP (°F) (p = 0.9188)			DECKTIME (Min) (p = 0.286)			TIME TO 50°F (min.) (p < 0.0001)			TIME TO 40°F (min.) (p < 0.0001)		
	n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD
BE	19	94.2	38.7	19	74.7	7.7	19	18.2	5.6	19	397	236.8	19	851	327.4
YF	15	81.4	12.2	15	74.8	7.0	15	15.9	5.4	12	352	142.8	12	786	113.8
AL	20	60.0	10.6	20	73.3	8.4	20	15.1	11.2	20	168	145.6	20	512	238.3
SM	15	66.5	17.7	15	75.8	5.8	15	14.0	5.9	14	218	100.9	14	410	160.7
BM	1	369.0		1	74.0		1	30.0		1	855		1	1435	
MM	10	28.2	7.8	10	76.1	6.4	10	18.6	10.5	9	179	45.9	9	313	80.1
ALL	80	73.2	44.6	80	74.8	7.1	80	16.4	8.2	75	275	195.7	75	611	312.8

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

Table 9b. Verification of post-harvest fish handling during longline fishing sets: fish temperature after 6 and 24 hours, quality grades (No. 1-5), total fish hold time and histamine concentration.

FISH	TEMP AT 6 HR (°F) (p < 0.0001)			TEMP AT 24 HR (°F) (p < 0.0001)			GRADE (p = 0.0132)			FISH HOLD TIME (min.) (p = 0.0032)			HISTAMINE (mg/100g) (p = 0.2806)		
	n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD
BE	19	51.2	6.8	19	35.6	3.6	19	2.1	0.8	19	13085	7733.6	19	0.35	0.22
YF	12	50.2	4.7	12	33.5	0.7	12	2.4	0.7	12	17502	5885.5	12	0.25	0.15
AL	20	44.0	5.8	20	32.4	0.7	20	1.9	0.3	20	10651	4789.9	20	0.21	0.15
SM	14	42.6	4.7	14	31.5	0.5	14	1.6	0.5	14	9507	4609.4	14	0.21	0.22
BM	1	65.2		1	40.1		1	2.0		1	21727		1	0.14	
MM	9	38.4	3.5	9	31.4	0.3	9	2.1	0.3	9	17110	7463.9	9	0.30	0.26
ALL	75	46.2	7.3	75	33.2	2.6	75	2.0	0.6	75	13073	6746.0	75	0.26	0.20

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

The mean fish weights were significantly different across species. This is only a reflection of the inherent differences between the fish species. However, fish weight was expected to have an effect on heat transfer rates reflected by differences in time and temperature parameters.

The mean initial core temperatures did not differ across the species and all species groups essentially began the on-board chilling process at similar starting temperatures. No difference was found in the mean deck time across species indicating that fishers tend to apply standard procedures in pre-processing fish before placing them in the fish hold.

Significant differences were found in the time and temperature parameters indicating that the differences in fish weight and physical differences in conformation between species has an effect on total heat load and heat transfer rates.

The total fish hold time differed significantly across species. The means ranged from a low of 6.6 days for striped marlin to a high of 15 days for the blue marlin. Fish hold time was compared to determine if the length of the time the fish was in the ice might have an effect on histamine accumulation.

After unloading, the fish were graded for quality and muscle samples were collected for histamine analysis. The mean quality grades differed significantly across species, however, no odor rejects (Grade 5) fish were found among these fish. Histamine concentration did not differ across species and the average histamine concentration for all of the fish in this sample set was 0.26 mg/100g (range 0.02 – 0.88 mg/100g, SD = 0.2mg/100g), well within the FDA defect action limit of 5 mg/100g. No histamine rejects were found within this sample set.

The post-harvest handling procedures and performance in temperature control documented during the 21 longline sets monitored during the project are considered representative of the Hawaii longline fleet that targets tuna. Those vessels that target swordfish tend to take longer trips and with the extended fish hold times, are known to produce high as well as low quality fish.

Did fish size make a difference?

Fish size logically makes a difference in chilling rates. The mean chilling rates reported in Tables 9a and 9b were ranked for comparison to illustrate the relationship between fish size (round weight) and chilling rates, where significant differences occurred between species (Table 10). The blue marlin was the largest group by weight followed by bigeye, yellowfin, striped marlin, albacore and mahimahi. If chilling rates are directly influenced by fish size, then similar species rankings should be expected for these time and temperature parameters.

Table 10. Comparison of pelagic fish ranked by mean fish size (weight) and chilling rates.

Weight	Initial Core Temp	Time to 50° F	Time to 40° F	Temp at 6 hours	Temp at 24 hours
BM (large) BE YF SM AL MM (small)	No statistical Difference (P>0.05)	BM (long) BE YF SM MM AL (short)	BM (long) BE YF AL SM MM (short)	BM (high) BE YF AL SM MM (low)	BM (high) BE YF AL SM MM (low)

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

The initial core body temperatures of these fish were not significantly different and ranged from 73.3 to 76.1° F (Table 9a). Ranking the time and temperature parameter means by species reveals that the blue marlin with the greatest size also had the

slowest chilling rate followed by bigeye and yellowfin tuna respectively. The last three fish in each category varied in order with mahimahi being the smallest fish by weight and having the fastest chill rate to 40° F and the lowest temperatures after 6 and 24 hours. This is also an indication of the size or weight factor. Heat transfer in mahimahi may also be influenced by other anatomical differences (body conformation, thickness of skin, fat content, etc.). Albacore had the fastest time to 50° F presumably because this species also had the lowest initial core temperature (73.3° F).

Results Obj. 4. Verify the relationship between post-harvest fish handling procedures and histamine accumulation.

Were the FDA fish handling guidelines met?

Yes for fish that are brought aboard alive. Uncertain for fish that died on the line. The on-board handling time and temperature parameters were evaluated in Objective 3. On average, all three fleets (troll, handline and longline) were capable of chilling fish within the guidelines offered by FDA, once the fish were brought on-board. Table 11 summarizes the critical information presented in Tables 9a and 9b from both dead and live longline fish. The uncertainty of the time of death for fish that are retrieved dead makes the calculation of exact chilling rates impossible. However, once fish were brought on-board, chilling rates from the time of boarding were determined accurately. Disregarding the additional time dead fish spend on the line, on average, all species groups caught by longline gear met the 50° F and 40° F guidelines. Mahimahi, striped marlin and albacore met the 50° F guideline, bigeye and yellowfin were close to meeting the guideline and the blue marlin was far from meeting this time and temperature goal. The FDA (1998) recognizes that if fish are well handled on-board the harvest vessel, that fish may be able to safely withstand somewhat more exposure to elevated temperatures during the post-harvest period. This indicates a certain amount of flexibility in adhering to the handling guidelines.

Table 11. Histamine concentration and compliance with FDA guidelines for pelagic fish caught by longline (retrieved alive and dead).

Fish	Were the FDA handling Guidelines met?		Were fish below the FDA histamine DAL? Histamine <5mg/100g
	temp <50° F w/in 6 hrs.	temp <40° F w/in 24 hrs.	
Bigeye tuna	No, but close	Yes	Yes
Yellowfin tuna	No, but close	Yes	Yes
Albacore tuna	Yes	Yes	Yes
Striped marlin	Yes	Yes	Yes
Blue marlin	No	Yes	Yes
Mahimahi	Yes	Yes	Yes
All fish (mean)	Yes	Yes	Yes

Did the post-harvest handling procedures adequately control histamine?

Yes. All samples collected from fish (alive and dead) with known on-board temperature histories fell within acceptable limits for histamine. The mean histamine concentration was 0.26 mg/100g (range = 0.02 - 0.88 mg/100g, SD = 0.20). The conclusion is that the

on-board handling methods observed and documented on Hawaii longliners are capable of adequately controlling histamine accumulation.

The group of longline fish was divided into fish that died and those that survived until being brought on-board. A comparison was made to determine if the mean histamine concentration differed between fish retrieved alive and those that died on the line. The results displayed in Table 12 indicate that there is no significant difference ($P>0.2$). This finding reduces the uncertainty about the adequacy of the current practices in preventing histamine accumulation in Hawaii longline fish that die on the line.

Table 12. Comparison of the histamine concentration of longline-caught fish retrieved alive and dead.

Fish	Histamine (mg/100g) Live fish at retrieval					Histamine (mg/100g) Dead fish at retrieval					Prob.
	N	mean	SD	min	max	N	mean	SD	min	max	
BE	14	0.38	0.23	0.04	0.72	5	0.24	0.19	0.02	0.53	P>0.5
YF	6	0.29	0.16	0.02	0.52	6	0.21	0.11	0.02	0.31	P>0.5
AL	10	0.18	0.13	0.02	0.39	10	0.23	0.17	0.02	0.53	P>0.5
SM	4	0.21	0.16	0.05	0.36	10	0.21	0.25	0.02	0.88	P>0.2
BM	0					1	0.14				
MM	7	0.33	0.28	0.02	0.74	2			0.05	0.26	
ALL	41	0.30	0.21	0.02	0.74	34	0.21	0.18	0.02	0.88	P>0.5

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

The laboratory maintained strict QA/QC procedures using histamine-spiked samples during each batch of fish run for histamine analysis. The average percentage recovery was 95.7% (range 89.2 - 109.0%, SD = 4.20%). The QA/QC for histamine analysis was deemed to be adequate and the histamine results are considered reliable.

Results Obj. 5. Determine the importance of fish quality grades and odors of decomposition as indicators of histamine concentration.

In addition to the fish sampled with known on-board temperature histories from studies in Objectives 3 and 4, fish were also sampled at the 2 fish auctions to represent fish delivered by the primary producer to the primary processor in the Hawaii fresh fish market. Fish were sampled from 42 commercial longline trips, 45 trolling trips and 32 handline trips. Fish were sampled between July 1998 and October 1999 during all four quarters of the year. This collection period encompassed the summer peak of fishing activity by all three gear types as well as the winter months when bigeye tuna are more prevalent.

The entire market sample set is displayed in Table 13. A total of 583 mixed pelagic fish were sampled, weighed, graded for quality and subjected to sensory evaluation for odors of decomposition and analyzed for histamine concentration. The sampling protocol attempted to collect equal numbers of fish in the 5 quality grades (Grades 1 – 4 and Grade 5 or "odor rejects"). Of the total, 119 fish were graded as odor rejects due to the presence of odors of decomposition detected by sensory examination.

Table 13. Histamine concentrations of commercial pelagic fish landed in Hawaii fresh fish market by gear type, fish species, weight, quality grade and sensory evaluation.

Gear type with common names	Round weight (lbs)				Grade 1 Histamine (mg/100 g)				Grade 2 Histamine (mg/100 g)				Grade 3 Histamine (mg/100 g)				Grade 4 Histamine (mg/100 g)				Reject Histamine (mg/100 g)								
	N	Mean	Min	Max	N	Mean	± SD	Min	Max	N	Mean	± SD	Min	Max	N	Mean	± SD	Min	Max	N	Mean	± SD	Min	Max					
Longline																													
Bigeye tuna	96	102	39	210	17	0.30	0.29	0.02	1.18	17	0.24	0.16	0.02	0.59	18	0.45	0.86	0.02	3.92	16	0.20	0.14	0.02	0.46	28	10.27	36.71	0.02	196.00
Yellowfin tuna	90	95	31	150	17	0.36	0.27	0.02	0.84	23	0.37	0.26	0.02	1.14	23	0.30	0.24	0.02	1.00	14	0.54	0.54	0.10	2.27	13	19.56	47.59	0.02	179.00
Albacore tuna	54	54	29	74	19	0.12	0.14	0.02	0.50	20	0.20	0.15	0.02	0.53						15	3.13	4.85	0.02	15.90					
Striped marlin	40	69	31	126	15	0.35	0.40	0.02	1.63	15	0.16	0.13	0.02	0.42						10	0.29	0.44	0.02	1.54					
Pacific blue marlin	30	179	99	429	15	0.29	0.49	0.02	2.02	15	0.33	0.44	0.02	1.72															
Mahimahi	85	19	5	45						31	0.32	0.51	0.02	2.91	1	0.69				53	0.39	0.93	0.02	5.74					
Handline																													
Bigeye tuna	4	51	31	107						4	0.48	0.19	0.21	0.70															
Yellowfin tuna	65	111	43	188						48	0.25	0.28	0.02	1.20	17	0.14	0.13	0.02	0.41										
Albacore tuna	43	51	36	67						43	0.19	0.16	0.00	0.78															
Troll																													
Yellowfin tuna	35	124	84	171						17	0.39	0.37	0.02	1.30	18	0.16	0.19	0.02	0.63										
Pacific blue marlin	24	246	103	599	18	0.14	0.21	0.02	0.72	6	0.11	0.11	0.02	0.33															
Mahimahi	17	18	8	34	17	0.20	0.25	0.02	0.92																				
All gear types																													
Bigeye tuna	100	100	31	150	17	0.30	0.29	0.02	1.18	21	0.28	0.19	0.02	0.70	18	0.45	0.86	0.02	3.92	16	0.20	0.14	0.02	0.46	28	10.27	36.71	0.02	196.00
Yellowfin tuna	190	106	31	188	17	0.36	0.27	0.02	0.84	88	0.31	0.30	0.02	1.30	58	0.21	0.21	0.02	1.00	14	0.54	0.54	0.10	2.27	13	19.56	47.59	0.02	179.00
Albacore tuna	97	53	29	74	19	0.12	0.14	0.02	0.50	63	0.19	0.15	0.00	0.78						15	3.13	4.85	0.02	15.90					
Striped marlin	40	69	31	126	15	0.35	0.40	0.02	1.63	15	0.16	0.13	0.02	0.42						10	0.29	0.44	0.02	1.54					
Pacific blue marlin	54	208	99	599	33	0.21	0.37	0.02	2.02	21	0.27	0.39	0.02	1.72															
Mahimahi	102	19	5	45	17	0.20	0.25	0.02	0.92	31	0.32	0.51	0.02	2.91	1	0.69				53	0.39	0.93	0.02	5.74					

A comparison of fish weight, grade and histamine was made between species for all gears combined (Table 14) in the market sample. The 6 fish species in the market sample differed statistically by weight ($P < 0.0001$) as expected. Blue marlin was the largest species, followed by yellowfin, bigeye, striped marlin, albacore and mahimahi.

Table 14. Comparison of weight, grade and histamine concentration between pelagic fish species for all gears.

Fish	N	Weight (lb) ($P < 0.0001$)		Grade (No. 1-5) ($P < 0.0001$)		Histamine (mg/100g) ($P = 0.259$)	
		mean	SD	mean	SD	mean	SD
BE	100	100	44.0	3.17	1.47	3.10	20.04
YF	190	106	32.2	2.57	0.99	1.62	13.40
AL	97	53	10.8	2.27	1.24	0.63	2.20
SM	40	69	23.7	2.38	1.60	0.26	0.35
BM	54	208	128.6	1.39	0.49	0.23	0.38
MM	102	19	8.6	3.40	1.71	0.34	0.74

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

The 6 fish species differed in average quality grades ($P < 0.0001$) with Grade 1 being the highest quality and Grade 5 being a reject. Blue marlin had the highest average quality score (1.39) followed by albacore (2.27), striped marlin (2.38), yellowfin (2.57), bigeye (3.17) and mahimahi (3.40) the lowest quality score in the market sample collected. It should be stressed that this is merely an analysis of the quality of the fish in the market sample set that was designed to compare across grades and is not a reflection of the typical catch make-up and fish quality available in the Hawaii market.

The comparison of histamine concentration in the market sample set by fish species for all fishing gears was not significant ($p = 0.259$). This is due to the wide variance in histamine values in the odor reject category (Grade 5).

Does fish quality grading and sensory evaluation effectively screen out fish with high histamine risk?

Yes. Every fish that contained histamine greater than 5 mg/100g fell into the category of odor rejects (Grade 5) made up of fish rejected due to inferior quality and odors of decomposition. The conclusion is that standard fish quality grading and screening fish for odors of decomposition is effective in eliminating fish with high histamine content.

In another study, sensory evaluation was shown to be highly correlated with histamine concentration in studies of histamine formation and decomposition at elevated temperature in mahimahi in Hawaii (Baranowski, et al, 1990).

Was there any evidence of histamine accumulation in the fish that passed sensory evaluation?

Yes. Four (4) out of the 464 fish of Grade 1 through Grade 4 had histamine concentrations above 2.0 mg/100g but none exceeded 4.0 mg/100g. These fish had all passed sensory evaluation. One (1) blue marlin (Grade 1, 2.02 mg/100g), 1 bigeye tuna

(Grade 3, 3.92 mg/100g), 1 mahimahi (Grade 2, 2.91 mg/100g) and 1 yellowfin (Grade 4, 2.27 mg/100g) were found. Fish that exceed 2.0 mg/100g histamine raise questions about inadequate on-board fish handling. Assuming that problems with handling these muscle samples between collection and histamine analysis did not occur to account for the elevated histamine, we conclude that the occasional fish can be found with more than 2.0 mg/100g histamine after passing sensory evaluation. However, these fish were still within legal and safe limits for sale and consumption.

Clearly, these few fish if improperly handled as they pass through the subsequent processing and marketing channels may have resulted in further histamine accumulation. However, with the FDA HACCP program, all histamine forming fish should be kept below 40° F at all times to control histamine. The potential problem increases after these fish leave the jurisdiction of FDA HACCP and into the hands of retailers, restaurants and consumers. In order to control histamine in these outlying groups, education and training in proper fish handling is greatly needed.

Is there a statistical difference between Grades 1, 2, 3, 4 and odor rejects?

The histamine concentration of longline fish was compared by quality grade and by species (Table 15). Only the bigeye and yellowfin tuna had sufficient numbers of fish from each of the 5 quality grade categories. The longliners in Hawaii do not generally produce the full range of quality grades for all species of fish. Grade 4 and odor rejects are relatively uncommon. Longliners do not produce the high quality, Grade 1 mahimahi that are only available from trollers making day trips. Grades 1 and 2 predominate in albacore, striped marlin and blue marlin in longline catches, while Grades 3 and 4 and odor rejects in these species are uncommon. During the market sampling, no blue marlin odor rejects were detected.

A comparison was made of histamine concentration between quality grades within species for longline-caught fish. Significant differences were found for albacore (P=0.0022) and yellowfin (P=0.0245)(Table 15).

Table 15. Comparison of histamine concentration for longline caught fish by species and quality grade.

Fish	Probability	Grade 1			Grade 2			Grade 3			Grade 4			Rejects		
		Histamine (mg/100g)			Histamine (mg/100g)			Histamine (mg/100g)			Histamine (mg/100g)			Histamine (mg/100g)		
		n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD	n	mean	SD
BE	0.3214	17	0.304	0.302	17	0.236	0.167	18	0.449	0.881	16	0.201	0.145	28	10.271	37.387
YF	0.0245	17	0.359	0.275	23	0.365	0.262	23	0.298	0.243	14	0.541	0.561	13	19.561	49.532
AL	0.0022	19	0.125	0.139	20	0.199	0.157							15	3.13	5.019
SM	0.3559	15	0.346	0.419	15	0.161	0.137							10	0.291	0.461
BM	0.8268	15	0.291	0.505	15	0.329	0.452									
MM	0.8682				31	0.323	0.519	1	0.690					53	0.388	0.942
ALL	0.0346	83	0.279	0.344	121	0.279	0.345	42	0.372	0.601	30	0.359	0.426	119	5.416	24.790

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna, AL = albacore tuna, SM = striped marlin, BM = Pacific blue marlin, MM = mahimahi)

Comparisons of mean histamine concentrations between grades within each species were made. In the albacore group, odor rejects had a greater average histamine (3.13 mg/100g) and differed from both Grade 1 (0.13 mg/100g, P= 0.0017) and Grade 2 (0.20 mg 100g, P=0.002) (Table 16). Within the yellowfin group, odor rejects had a mean histamine concentration of 19.56 mg/100g and differed from Grade 1 (P=0.0063),

Grade 2 (P=0.0038), Grade 3 (P=0.0037) and Grade 4 (P=0.0095). Grades 1 through 4 however, were not significantly different from each other.

Table 16. Comparison and ranking of mean histamine concentrations by grade for longline caught fish (Reject = Grade 5).

Fish	Grade vs. Grade comparison	Probability	Rank of Means	Histamine (mg/100g)
Albacore	Grade 1 vs. Reject	0.0017	Reject	3.13
	Grade 2 vs. Reject	0.002	Grade 2	0.20
Yellowfin	Grade 1 vs. Reject	0.0063	Grade 1	0.13
	Grade 2 vs. Reject	0.0038	Reject	19.56
	Grade 3 vs. Reject	0.0037	Grade 4	0.54
	Grade 4 vs. Reject	0.0095	Grade 2	0.37
All species	Grade 1 vs. Reject	0.013	Grade 1	0.36
	Grade 2 vs. Reject	0.006	Grade 3	0.30
	Grade 3 vs. Reject	0.052	Grade 4	0.35
	Grade 4 vs. Reject	0.087	Grade 2	0.28
			Grade 1	0.28

When all species were considered, comparisons of the mean histamine concentrations of odor rejects and the other quality grades were significant except for Grade 4.

By combining the Grades 1 through 4, the mean histamine concentrations of acceptable quality fish and odor rejects were compared (Table 17). In this way, the odors of decomposition were evaluated as indicators of histamine risk. Bigeye and yellowfin tuna were the two species with sufficient numbers of Grades 1 through 4 and odor rejects (Grade 5) needed to make the following comparison. For both bigeye and yellowfin tuna, odor rejects had significantly higher mean histamine concentrations than fish without odors of decomposition (Grades 1 - 4 combined). The mean histamine concentration for all of the acceptable quality bigeye and yellowfin (Grades 1 - 4) was 0.30 and 0.37 mg/100g respectively, well below the defect action level. By contrast, bigeye and yellowfin odor rejects had mean histamine concentrations of 10.27 and 19.56 mg/100g respectively, far exceeding the defect action limit.

Table 17. Comparison of histamine between grade 1 through 4 combined and odor rejects (Grade 5) longline caught bigeye (BE) and yellowfin tuna (YF).

Fish	Probability	Grades 1 to 4 combined. histamine (mg/100g)			Grade 5 (odor rejects) Histamine (mg/100g)		
		N	mean	SD	N	mean	SD
BE	0.0292	68	0.30	0.48	28	10.27	37.38
YF	0.0007	77	0.37	0.33	13	19.56	49.53

(Abbreviations: BE = bigeye tuna, YF = yellowfin tuna)

What can be said about the odor reject category?

Table 18 displays the numbers and percentages of the odor rejects separated by species, that had histamine levels of 0 - 1.99, 2.00 - 4.99 and >5.00 mg/100g. These

values were used because of the regulatory implications. The defect action level for histamine in seafood is 5 mg/100g. Histamine concentration between 2.00 and 4.99 mg/100g although acceptable, is cause for concern because it indicates marginal post-harvest handling. Fish with less than 1.99 mg/100gm are acceptable for sale.

A total of 119 fish rejected from the market due to odors of decomposition (Grade 5) were sampled during the study in order to achieve close to equal numbers of fish to represent each of the 5 grade categories in the market sample.

All of the histamine rejects found in the study were initially rejected for decomposition. There were only 14 fish found that exceeded the histamine defect action limit (5mg/100g) amounting to only 11.7% of the 119 odor rejects. The majority (85%) of the decomposed fish had low and acceptable levels of histamine (<1.99mg/100g). Only 4 fish (3.3%) were found with histamine concentrations between 2.00 and 4.99 mg/100g.

Table 18. Histamine concentrations of longline odor rejects.

Fish	Odor Rejects N	Histamine		
		<1.99mg/100g N (%)	2 - 4.99mg/100g N (%)	>5mg/100g N (%)
Bigeye	28	22 (78.5%)	1 (3.6%)	5 (17.9%)
Yellowfin	13	8 (61.5%)	1 (7.7%)	4 (30.8%)
Albacore	15	10 (66.6%)	1 (6.6%)	4 (26.6%)
Striped Marlin	10	10 (100%)		
Blue Marlin	0			
Mahimahi	53	51 (96.2%)	1 (1.9%)	1 (1.9%)
TOTAL	119	101 (85%)	4 (3.3%)	14 (11.7%)

Five (5) of the bigeye tuna odor rejects (17.9%) contained histamine levels above 5 mg/100g. Four (4) of the yellowfin odor rejects (30.8%) were also histamine rejects. There were 4 albacore histamine rejects (26.6%) among the 15 albacore odor rejects. The striped marlin odor rejects were all within acceptable limits for histamine. No blue marlin odor rejects were found during the market sampling. Only 1 mahimahi histamine reject (1.9%) was found among the 53 mahimahi odor rejects. This finding is of great interest in that mahimahi is one of the two most commonly implicated species in cases of histamine poisoning in Hawaii. It may be that the domestically produced mahimahi reported to cause histamine poisoning in Hawaii are not landed by longline vessels. More thorough epidemiological reporting and investigations would be required to answer this question.

What was the source of the odor rejects and high histamine fish?

All of the odor rejects were from longline sets and none were found in the landings of Hawaii trollers and handliners during the study. The lack of rejects from trollers and handliners is likely due to the short period to time between death and delivery to the primary processor. Evaluating the histamine accumulation that may occur after delivery to the market was not one of the objectives of this study.

Longline trips in Hawaii can be categorized by target species into tuna and swordfish trips. Fishing trips targeting swordfish and tuna differ in primary target species, some

aspects of the fishing method, location of fishing effort and especially in the trip length. Swordfish trips also harvest tuna and the associated pelagic fish species. For each of the odor rejects, the type of longline trip taken (targeting tuna or swordfish) and the trip length were evaluated. Table 19 presents the number of trips, the number of vessels, the type of fishing trip by targeted species and the total length of the trips in days for vessels that produced the odor rejects sampled during the study.

Table 19. Trip length for vessels producing fish rejected from the market for odors of decomposition and exceeding the FDA histamine defect action limit.

Fish	Type of trip	mean (days)	min (days)	max (days)	SD (days)	No. of trips	No. of vessels
Odor rejects	All trips	28.84	11	51	11.24	19	16
	Tuna trips	19.00	11	29	6.03	6	
	Swordfish trips	33.38	20	51	10.13	13	
High histamine rejects	All trips	31.67	11	50	11.51	9	9
	Tuna trips	19.33	11	29	9.07	3	
	Swordfish trips	37.83	25	50	8.28	6	

The odor rejects sampled during this study came from 16 different longline vessels and 19 different fishing trips. The majority of the odor reject fish were collected from fishing trips in which swordfish were targeted (13 out of 19 trips). Due to the longer distances traveled during swordfish trips, trip length (33 days) tended to be longer than for trips targeting tuna (19 days). The trip length typically reported for tuna trips in Hawaii is 14 to 21 days and for trips that target swordfish, the trip length is considerably longer, from 30 to 45 days (WPRFMC, 1995).

Product shelf life and decomposition are related to storage time, temperature control and sanitation. No odor rejects were found in troll and handline vessel landings presumably due to the on-board handling methods observed and the relatively short storage time (<2 days). There were more odor rejects sampled from swordfish trips than from tuna trips. Swordfish trips may have exceeded the upper limits of the storage period for chilled tuna and associated pelagic fish, allowing ample time for decomposition to occur, even when the fish were handled properly on-board.

Was there a difference in histamine accumulation between tuna and swordfish trips?

The mean histamine concentration for all of the histamine rejects was 41.05 mg/100g (Table 20). Ten (10) out of the 14 histamine rejects came from swordfish trips with a mean concentration of 34.56 mg/100g. These histamine rejects were produced on 6 different fishing trips targeting swordfish ranging in length from 25 to 50 days with a mean of 37.8 days (Table 19).

Four (4) out of 14 histamine rejects came from fishing trips targeting tuna (Table 20). The mean histamine concentration of these fish was 64.84 mg/100g, twice the amount found in histamine rejects from swordfish trips. These histamine rejects came from 3 different vessels, which made trips ranging from 11 to 29 days with a mean of 19.33 days (Table 19).

Table 20. The histamine concentration of histamine reject fish by the type of longline fishing trips.

Type of longline fishing trip producing histamine rejects	Number of histamine reject fish	Histamine concentration (mg/100g)		
		mean	range	SD
All trips	14	41.05	5.33 – 196.00	63.25
Swordfish trips	10	34.56	5.33 – 196.00	55.04
Tuna trips	4	64.84	5.74 – 179.00	98.88
Single problem vessel	5	88.98	5.33 – 196.00	91.26
Swordfish trips w/o the problem vessel	7	16.33	8.09 – 39.60	10.84
Tuna trips w/o the problem vessel	2		5.74 and 9.79	

Closer analysis revealed that a single vessel had caused 5 of the histamine reject fish on 2 separate fishing trips (Table 20). The mean histamine concentration of these fish was 88.98 mg/100g. This vessel produced the fish with the three highest histamine concentrations (45.3, 179.0 and 196.0 mg/100g) found in the study. This vessel had made a tuna trip of 18 days and a swordfish trip of 25 days. The trip lengths were not excessively long by fleet comparison. This indicates that there may be a problem on this particular vessel with a possible lack of awareness or understanding of histamine formation, proper fish handling, storage and sanitation.

After removing the fish produced by this single problem vessel from the calculations, the mean histamine concentration of histamine rejects from swordfish trips was 16.33 mg/100g (Table 20). The two histamine rejects from the remaining tuna trips contained 5.74 and 9.79 mg/100g histamine.

The single mahimahi histamine reject found in the market survey (Table 18) was sampled from a tuna vessel after only 11 days at sea. This single fish had a histamine concentration of 5.74 mg/100g, just above the defect action limit. The relatively short trip length indicates that there may have been fish handling problems on this vessel during the trip that would have allowed odor rejects and a histamine reject to form during the relatively short trip. Other possible reasons for this finding may have been related to the length of the longline gear, the soak time, depth of the gear, etc.

The odor reject and histamine reject data allow for identification of problem vessels and also point to a basic problem with the relatively long duration of longline trips targeting swordfish. In efforts to reduce the histamine risk in the fresh fish landings in Hawaii, reject data could be used on a routine basis to target training efforts on problem vessels to help them to improve their handling practices and understanding of the histamine hazard. The regular collection and analysis of reject data could also be used overtime to determine the maximum trip length for fresh tuna and associated pelagic fish stored in ice. This information could be valuable in making recommendations to vessel operators on how to reduce the volume of decomposed fish and economic discards.

Estimating the prevalence of high histamine fish in the Hawaii fresh fish landings.

As discussed, the number of odor rejects and histamine rejects evaluated in this study was artificially inflated as a result of the study design and sampling protocol. In this study, 20% of the fish sampled from the market were Grade 5 or odor rejects in order to compare quality and sensory attributes as indicators of histamine risk across the 5 Grade categories.

The actual rejection rate for decomposition is much less. The Honolulu Fish Auction estimates that the annual percentage of odor rejects in the fish landings is less than 0.01%. Using this estimate in combination with the percentage of histamine rejects (11.7%) within the odor reject category found in this study, a low frequency rate of 0.00117% for high histamine fish is estimated for the fresh market landings in Hawaii.

With this extremely low prevalence of high histamine fish in the fresh fish landings, it is difficult to see how lot sampling and histamine testing would be of any value in screening out high histamine fish. Assuming that the odor reject category contains all of the high histamine fish, as was found in this study, the rationale for random lot sampling and testing for histamine is highly questionable.

HACCP is not a zero-risk system and should address "*likely hazards*". Elevated histamine concentration does not appear to be likely to occur in fresh pelagic fish caught by hook and line gear that fall within acceptable quality grades and pass sensory evaluation for odors of decomposition. Applying lot sampling and histamine testing to the fresh pelagic fish landings in Hawaii is therefore deemed inappropriate and unwarranted, based on the best available scientific data and understanding of industry practices.

Results Obj. 6. Develop a HACCP-based strategy for the control of histamine for the fresh tuna industry.

The results of Objectives 1 through 5 have served to describe the histamine hazard in the Hawaii fishery, first by focusing on the epidemiological evidence on the pelagic fish species produced and landed by the troll, handline and longline fleets. Post-harvest fish handling procedures on commercial fishing vessels were also monitored and documented at sea to determine the performance of the vessels in meeting the FDA time and temperature guidelines for the prevention of histamine accumulation. The handling procedures documented during commercial fishing trips demonstrated that these vessels were capable of effectively control histamine accumulation in the fish. Lastly, fish quality grading and sensory evaluation were shown to be effective methods for culling out all fish with histamine concentrations above the defect action limit.

The application of these findings to develop a rationalized HACCP-based approach to controlling histamine in the fresh tuna industry is the remaining task. A practical and effective HACCP-based system is required for the important interchange between the fishing vessels (primary producer) and the primary processor in the processing and marketing chain. The primary processor takes the responsibility for extending the reach of HACCP to the fishing vessels at sea where histamine problems are most effectively controlled.

It has been argued that lot sampling and testing for histamine is not an effective HACCP-based method for histamine control in hook and line fisheries. Problems with obtaining detailed on-board handling data for individual fish at sea have been discussed. An alternative approach is recommended that integrates scientific and industry knowledge

of histamine accumulation and standard industry practices, specific to the Hawaii pelagic fishery.

The Hawaii VSOP Approach for histamine control.

The VSOP (vessel standard operating procedures) for post-harvest fish handling at sea is designed for Hawaii's fresh tuna fishery and integrates the FDA fish handling guidelines and new information generated during on-board research in this study. The VSOP details the minimum handling parameters required for the control of histamine accumulation in histamine-forming fish species (Figure 7). The guidelines recommended by FDA of chilling fish to below 50° F within 6 hours and to below 40° F within 24 hours of boarding are used in the VSOP. Results of this study demonstrated that on average, Hawaii longline, handline and trolling vessels are capable of meeting these guidelines once the fish are brought aboard and adequately controlling histamine accumulation.

The primary processor requires some form of verification that vessel crews adhere to the VSOP during fishing trips. Primary processor should prepare VSOP documents that can be signed by the vessel owner and captain to be kept on file for each vessel, as a pre-requisite of purchasing or marketing agreements. The VSOP on file with the primary processor should be renewed annually. A letter of assurance (LOA) should be signed and submitted each time the vessel delivers fish to the market as a written guarantee of compliance with the VSOP (Figure 8) and the HACCP Plan of the receiving company.

In addition, the adequacy of the icing in the hold should be checked at the time of unloading. The amount of ice at the beginning of the trip should be reported. The time the first and last fish were boarded is also very important information. Internal fish temperatures should be recorded for 3 fish per ton at the time the fish are delivered. All fish that are over 24 hours out of the water should be below 40° F. Fish out of the water between 6 and 24 hours should be less than 50° F at receiving. Fish out of the water less than 6 hours can be received at above 50° F as long as the icing is adequate.

It has been demonstrated that odors of decomposition are reliable indicators of fish with high histamine concentration in the Hawaii fresh tuna industry. As a vital part of the VSOP approach, individual fish should undergo quality grading and sensory evaluation to screen out fish with odors of decomposition at the time of delivery. This standard industry practice in Hawaii is now incorporated into a practical HACCP approach. Quality grading and sensory evaluation are conducted by the HACCP manager or scale master, again by the auctioneer and finally by the individual buyers at the point of purchase. This redundancy of quality and sensory checks ensures that decomposed, along with high histamine-risk fish are effectively culled from the market. Decomposed fish should be rejected on an individual fish basis and have no impact on the rest of the fish in the load as in the lot sampling methods.

Hawaii's two display auctions adopted a VSOP system soon after the FDA HACCP program became effective in December of 1997 (Kaneko, 1997a,b,c). Cooperation from fishers has been exemplary to date. The VSOP has focused attention to on-board handling details, increasing the awareness of fishers to their responsibility in providing high quality and safe fish to the market. The VSOP approach integrated in-depth knowledge of standard industry practices with available scientific understanding of histamine formation and the principles of HACCP. The current study was proposed and

conducted to provide scientific validation of the efficacy of the Hawaii VSOP system for controlling histamine accumulation in tuna and associated pelagic fish.

Figure 7. Vessel Standard Operating Procedure (VSOP) document.

COMPANY X, Inc
VSOP
(Vessel Standard Operating Procedures)
On-board Fish Handling for Histamine Control

Fishing Vessel: _____

Owner: (print) _____ (sign) _____ (date _____)

Captain: (print) _____ (sign) _____ (date _____)

This is to verify that the following standard operating procedures for on-board fish handling are practiced on this vessel and that any significant deviation from these practices will be noted and the receiver notified prior to unloading. This VSOP is submitted in cooperation with the receiver's HACCP Program designed especially for the prevention of histamine accumulation in susceptible fish species.

Fishing Method: longline / handline / troll

Refrigeration Method: ice slurry / ice alone / ice with refrigerated fish hold / RSW

Sanitation:

The fish holds are cleaned and sanitized after each trip using a dilute chlorine bleach solution (specifically, sodium hypochlorite solution of 100 ppm). Clean, new ice made from potable water is loaded into the fish hold at the start of each fishing trip. Fish holds are not used to store fuel. Fish holds are kept free of chemicals and lubricants used on-board the vessel.

Fish Handling Method:

Fish are handled carefully, kept clean and chilled rapidly in order to prevent the potential formation of histamine in susceptible fish species.

Fish are landed individually by hook and line, gaffed and immediately stunned with a club and bled using gill and tail cuts. The fish is rinsed with clean seawater and placed immediately into ice or ice slurry. Fish may be kept whole, gilled and gutted or headed and gutted. This process takes no more than 15 minutes from the time of boarding.

Fish are chilled to an internal temperature of 50°F within 6 hours of boarding. Fish temperatures are brought down to < 40°F within a total of 24 hours of boarding. Fish are kept properly iced during storage on-board the vessel to maintain fish temperature <40°F until unloading.

Figure 8. Letter of Assurance (LOA) of compliance with VSOP.

COMPANY X, INC.
VESSEL HACCP RECORD

Letter of Assurance (LOA)

This certifies that the fish delivered to COMPANY X from the described fishing trip were handled in accordance with the current signed VSOP (vessel standard operating procedures) document on file at COMPANY X. This information is provided as a component of the COMPANY X VSOP program for compliance with US FDA HACCP regulations (21 CFR Part 123).

Vessel Name: _____

Fishing Method: (circle one) (longline / handline / troll)

Captain: (print) _____ (sign) _____ (date) _____

Trip details:

Date trip started:	_____	Time departed:	_____
Date first fish caught:	_____	Time fish caught:	_____
Date last fish caught:	_____	Time fish caught:	_____
Date of unloading:	_____	Time started:	_____

Last fish caught: () dead for more than 24 hours at delivery
 (check one) () dead for 12 to 24 hours at delivery
 () dead for less than 12 hours at delivery

Cooling Methods:

Tons or lbs. of ice at start of trip: _____

Icemaker capacity: _____/day

Refrigerated hold? (circle) (yes / no)

Tons or pounds of fish: _____ total estimate

***To be completed by COMPANY X:**

Icing adequate at time of unloading? (yes / no)

VSOP on file at COMPANY X? (yes / no)

Signature of COMPANY X staff: _____

The VSOP system designed for the Hawaii fresh tuna industry is tailored to the unique industry setting and fishery. The VSOP system currently in place is effective only because of the special relationship between the fleets and the market through the 2 fish auctions that receive the majority of the commercial fish landings in the state. The Hawaii fish auction system is unique in the US and allows each fish to be displayed, closely evaluated for quality and odors of decomposition and sold on an individual fish basis. An important feature is that buyers have no obligation to bid on or purchase fish

and must take the full responsibility to evaluate the fish that they buy for quality and relative safety indicators. The auction system rewards high quality with higher prices and links the interests of fishers with the auctions directly. Fishers recognize the direct relationship between proper handling, fish quality and monetary returns to the vessel. This emphasis on higher quality at the same time promotes the control of histamine. The VSOP system helps the auctions to encourage fishers to practice proper on-board fish handling methods that are known to prevent the accumulation of histamine.

Precautions about the application of the VSOP system.

The VSOP system is supported by the results of the studies designed and conducted to evaluate the specific industry and environmental conditions in Hawaii. Extrapolation of this information to other fisheries is not acceptable. The principles of HACCP require hazard analysis to be conducted for each operation and industry sector as the first step in developing a HACCP program.

To illustrate this point, the same information and VSOP approach to controlling histamine cannot be applied to frozen tuna products. This is especially true for carbon monoxide or filtered wood smoke treated frozen tuna. This is because the important characteristics of fresh tuna grades (muscle color and clarity) are altered in frozen tuna unless they are frozen to ultra-low temperatures (-50° F). Carbon monoxide treatment of tuna creates an unnatural and unusually stable red muscle color that does not have the normal characteristics of fresh untreated tuna. The use of carbon monoxide eliminates the ability to accurately judge the true product quality and therefore product safety. Most importantly, the use of carbon monoxide to treat tuna alters the odor characteristics of the product. This makes the critical step of sensory evaluation uncertain and unreliable in screening out fish with high histamine risk. This is in sharp contrast to the demonstrated efficacy of sensory evaluation in culling high histamine-risk fish in landings of chilled, untreated tuna and associated pelagic fish species in Hawaii.

Results Obj. 7. Communication of results to the FDA Office of Seafood.

Copies of the final report will be submitted to the FDA Office of Seafood. Dr. George Hoskin, Director for Science and Technology in the Office of Seafood served as the primary point of contact. He will distribute the report copies to specialists at the FDA.

- B. If significant problems developed which resulted in less than satisfactory results, they should be addressed.

No significant problems occurred. However, during the study, vessel-based research was conducted on-board commercial trollers, handliners and longliners. During the initial trips on trollers and handliners, the post-harvest fish handling methods were documented and it was determined that the cooling method used (ice slurry) was more efficient than the method used on longliners (ice alone). The catch rates on trollers and handliners also proved to be much lower than expected and data collection from these two fleets was inefficient. The decision was made to focus more attention on the longline fleet that produces the bulk of the commercial fish landings in Hawaii and could be studied in multiple productive longline sets more reliably than on troll or handline trips.

C. Description of need, if any, for additional work.

Follow-up work should focus on prevention of histamine problems in the fresh tuna industry. Additional work recommended includes:

- Monitor the efficacy of the VSOP system for controlling histamine risk by reviewing epidemiological data and auction company records. Assist State of Hawaii Department of Health investigators in thorough investigations of incidents of histamine poisoning.
- Prepare training materials on seafood safety, quality and proper handling for commercial fishers, fish auctions, fish processors, and retail and restaurant staff. Should be translated into Korean, Vietnamese, Chinese, Samoan and Filipino.
- Use reject data to target training and education efforts for vessels with quality, seafood safety and on-board handling problems. The appropriate training materials should be distributed to the vessel owner and captain each time the vessel delivers decomposed fish in a continuing effort to improve the safety, quality and value of fish landed in Hawaii.
- Work with fishers to develop alternative on-board fish handling and marketing strategies to reduce the likelihood of delivering decomposed fish, improve the economic viability of the vessel and reduce the histamine risk in the market.
- Conduct practical training workshops for commercial fishers on seafood quality, safety, VSOP and HACCP compliance. This might be incorporated as a requirement of the VSOP system for vessels supplying fish to the first receiver.
- Conduct practical training workshops for fish auctions, processors, wholesalers and distributors on seafood handling, seafood safety and the VSOP system for histamine control.
- Conduct practical training workshops for retailers and restaurant staff on proper seafood handling and seafood safety.
- Conduct practical training workshops for recreational and subsistence fishers about seafood safety and proper fish handling in an effort to reduce histamine poisoning from non-commercial channels.
- Conduct practical training workshops for FDA Inspectors on fresh tuna quality and the VSOP system to ensure a working understanding of industry practices, seafood safety controls and HACCP compliance.
- Investigate the source, fishing methods and cause of the high histamine risk in imported mahimahi. Considering how rapidly properly handled mahimahi can be chilled, the handling on-board fishing vessels supplying the US market with imported mahimahi must be grossly inadequate. Investigate practical methods for detecting high histamine concentration in frozen product.

- Further evaluate the relative risk of histamine accumulation in the Pacific blue marlin as this species is the largest and most difficult to properly chill. Develop practical measures to improve the chilling rates on small trolling vessels for properly handling large fish.
- Continue to study histamine accumulation, quality shelf life and the relative safety of fresh fish landed by troll, handline and longline vessels as they are processed and distributed through the market channels. The present study only focused on the histamine risk at the time of delivery from the vessels to the first receiver.

VII. Evaluation:

A. Describe:

1. Were the goals and objectives attained? How? If not why?

Objective 1. Evaluate epidemiological data on histamine poisoning in Hawaii.

Yes. Ten (10) years of epidemiological data on reported cases of histamine poisoning in Hawaii were reviewed and summarized. These data indicate that tuna and mahimahi are the most frequently implicated fish species. This information is helpful in identifying the other less common species implicated.

Objective 2. Develop Fleet on-board handling profiles (trollers, handliners and longliners).

Yes. Fleet profiles were written for the troll, handline and longline fleets. Fishing and fish handling practices were evaluated and summarized based on observations at sea, interviews with fishers and literature review.

Objective 3. Verify the post-harvest fish handling procedures.

Yes. On-board fish handling practices were observed and monitored during commercial fishing trips by the project researchers. Vessels representative of the three fishing gear types participated and provided an opportunity for verifying the fish handling procedures. Temperature loggers proved to be valuable in recording time and internal body temperature from the time the fish were brought aboard until they were unloaded at the end of the trip.

Objective 4. Verify the relationship between post-harvest fish handling procedures and histamine accumulation.

Yes. The same fish monitored at sea with the temperature loggers were also sampled for histamine at the time of unloading. This allowed for histamine analysis of fish with known time and temperature histories. This allowed for the verification of fish handling procedures in terms of controlling histamine accumulation.

Objective 5. Determine the importance of fish quality grades and odors of decomposition as indicators of histamine concentration.

Yes. Fish were sampled from the two fish auctions in Hawaii. Each fish was graded for quality and odors of decomposition. It was determined that within the 583 fish sampled,

a total of 14 high histamine fish were effectively isolated and culled from the market by using quality grading and sensory evaluation. Odors of decomposition were demonstrated to be practical and useful in culling fish with high histamine risk.

Objective 6. Develop a HACCP-based strategy for the control of histamine for the fresh tuna industry.

Yes. The VSOP strategy for controlling histamine in fresh tuna and associated species in Hawaii's fishery was developed to integrate industry practices and knowledge with the principles of HACCP and the best available scientific knowledge. The current project served to strengthen the understanding of the histamine risk in the Hawaii fishery and how our major fleets are able to control histamine accumulation. The VSOP system draws from the Vessel Records Approach presented by the FDA. It links fishers with the primary processors in a system of mutual responsibility to produce safe fish and to ensure that safe fish are sold into the market. The auction system adds an additional safeguard in that auction buyers are representatives of the secondary processors and have the opportunity during the display auction to carefully judge quality and safety of the fish they purchase. There is no obligation to buy.

Objective 7. Communication of results to the FDA Office of Seafood.

Yes. Draft copies of this report were sent to Dr. George Hoskin, Director of Science and Technology at the FDA Office of Seafood. He distributed copies to professional staff for comment. Detailed comments on the draft were received from Robert Samuels of the FDA Program and Enforcement Branch. The comments were considered and many of them were helpful in strengthening this final draft. The FDA Office of Seafood will receive copies of the final report. It is anticipated that the discussion with the FDA about the validity of the Hawaii VSOP system for controlling histamine in fish will include detailed evaluation of the findings of this project. It is hoped that this research funded by NOAA through the Saltonstall-Kennedy Fisheries Research Program will contribute to the sustainability of the Hawaii fresh tuna industry by streamlining food safety assurance efforts, marrying effective industry practices with the science of histamine controls and the principles of HACCP.

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