The Effect of Operational Procedures on Fish Grown in Recirculating Aquaculture Systems

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Introduction

While a great portion of aquaculture research has centered on the growing of fish in open and recirculating systems, a limited amount of studies have focused on the various production and processing methods that could be employed when fish are ready for slaughter. While harvesting wild species at sea imposes limits based on the capabilities of the fishing vessel, domestically produced fishery products provide the opportunity to introduce a variety of killing, processing, and storage procedures that ensure the delivery of a high quality product to the marketplace.

The objectives of this series of studies was to examine various operational methods affecting slaughter and storage that could affect product quality as well as overall consumer acceptance.

A slaughter study focusing on the effects of kill method, stress, bleeding, and rigor on hybrid striped bass was conducted at a commercial aquaculture facility. The purpose of this study was to relate methods of sacrifice (cervical dislocation, electroshock, CO₂, ice chill, anesthetization, bleed, and lowered pH) to percent yield, proximate analysis, elemental analysis, and textural properties.

A second independent bleeding study to determine the effect of bleeding on color and sensory attributes was performed on hybrid striped bass at a University aquaculture facility. In this experiment a control group (no bleed) was compared to fish bled by gill bleed after 0, 60, 90, and 120 minutes.

Material and Methods

Killing Method Comparison

Prior to killing, fish were divided into two groups representing stressed (S) and not stressed (NS). Agitation was used to introduce stress on the first group. During processing, these two groups were equally subdivided into groups representing bled and unbled fish. The fish were bled by a
cut near the gills through an artery. The groups were identified as stressed bled (SH), stressed unbled (S), non-stressed bled (NSH), and non-stressed unbled (NS). Fish representing each of the stressed and bled groups were killed using each of the following methods.

1. Control - cervical dislocation (C)
2. Electroshock (E)
3. Carbon Dioxide (CO\(_2\)), pH 4.5
4. Rapid chilling with ice (FI)
5. Slow chilling with ice (SI)
6. Anesthetize with MS222 (MS)
7. Bleeding from head and tail (HT)
8. Lower pH with simultaneous O\(_2\) and CO\(_2\)

Half of the fish in each group were left whole and stored at 0°C for five days. The other half were processed immediately. A single fileter processed all samples representing each of the sampling groups in order to minimize cutting variation for the percent yield data.

**Proximate Analysis**

Samples were dried at 100°C overnight (~18 hr), stored in a desiccator, ground to a powder and tested for crude protein (AOAC 24.028-24.040), acid detergent fiber (ADF) (AOAC 7.7074 and lipids (ether extract, AOAC 24.005).

**Elemental Analysis**

Samples were dried at 100°C overnight (~18 hr) and ground to a powder. One gram of powder was weighed and digested in nitric acid. The digestate was then diluted to 100 mL with distilled water and filtered through a Whatman No. 1 filter to remove particulate matter. Analysis for all elements, excluding Arsenic (As), Mercury (Hg), Lead (Pb), and Selenium (Se) were run simultaneously on a Jarrell-Ash ICP 9000 Inductively Coupled Plasma Spectrophotometer (ICP). The As, Hg, and Se samples were run sequentially on a Jarrell-Ash Atom Scan 2400 ICP.

**Texture Analysis**

Samples were tested three days after harvest since that was thought to be a representative time between sacrifice and delivery to market. Samples measuring 4 cm x 2 cm were analyzed using the Instron Model 1011, equipped with a L.E.E. Kramer shear compression cell and 500 kg load transducer. Total energy (newton meter/kg fish) and peak force (newtons/kg fish) were calculated from texture profiles obtained.

**Timed Bleeding Study**

In this experiment, groups of fish were harvested, placed live on ice and bled by gill bleed after a predetermined interval (0, 60, 90, 120 min). A no bleed experimental group served as the control.
Fish were allowed to bleed for 10 min., then rinsed and packed on ice until analyses. After 24 hr. fish was fileted and weights determined for yield data. Filets representing each group were oven baked at 350°F for 20 min. Whole, raw filet, and cooked filet were analyzed for color differences using a Minolta Chroma-meter. L*, a*, and b* values were used to measure color differences. Sensory characteristics (appearance, redness, vein prevalence, odor) were evaluated by an experienced panel of ten departmental personnel. Taste and texture were rated on the cooked filet only. A preference score was also included to determine market success.

Results and Discussion

Average percent yield differences for pre and post-rigor analysis are shown in Table 1. Statistical analysis (SAS System, Cary, NC) showed that percent yield was significantly affected (p<0.05) by processing the fish post-rigor. On average, a 1 to 5 percent increase in processing yield was revealed by this study. Killing method and bleeding did not seem to have an effect on yield data.

Proximate analysis data (Table 2) revealed no major differences between treatment groups. Percent protein values were relatively similar in all groups and ranged from 78.8 to 91.2 percent. More variable results were seen in the ADF and lipid analysis. Further study in this area is recommended to determine if any dissimilarities may exist.

The elemental study analyzed filets for P, K, Ca, Mg, Na, S, Zn, Mn, Cu, Fe, Al, B, Cd, and Ni. Except for boron and zinc, no major differences were observed. It is not known why kill method or bleeding would affect these two particular elements. None of the samples tested showed element concentrations that could impact public health or exceed state and federal tolerance levels.

Texture analysis was performed on the raw as well as the cooked samples. Graphs of the entire shear/compression process of each sample was linearly regressed to provide an indication of tenderness. Total energy and peak force were calculated from the texture profiles. Statistical analysis showed that texture was not significantly affected (p>0.05) by the killing method, however the data revealed that rigor significantly (p<0.05) affected the texture of the fish filet. Fish completing the rigor mortis stage had filets much tougher than the filet cut before the onset of rigor. Post mortem conditions occur when the muscle becomes rigid due to the disappearance of adenosine triphosphate (ATP). Actin and myosin which comprise the main components of muscle tissue will become bound when the ATP bridge connection is eliminated. The condition occurs when glycogen is depleted and glycolytic pathways producing ATP stop. Therefore, any condition that depletes glycogen reserves will decrease time to rigor. Stress also significantly (p<0.05) toughened the filet as was demonstrated by a higher peak force and total energy than the non-stressed. Stressed fish tend to expend more energy swimming or struggling, reducing the muscle glycogen storage, thereby shortening time to rigor. Two additional methods used to increase time to rigor are low pH and slow cooling, however, the results in this study were contrary to this effect. It is possible the lowered pH may have introduced stress to the fish and caused a toughening effect on the filet.
The second bleeding study incorporating bleed time as a variable showed a significant difference (p<0.05) between 0 time bleed and no bleed samples in appearance, redness, and preference. Panelists consistently scored appearance (Figure 1) as lighter on the 0 time bled fish. Redness was not noticed as much by the panel for the 0 time while the no bled fish was given a higher rating in this category. In addition, the panel perceived the redness as bruising thereby lowering the preference scores for that fish. Other bleeding times versus the no bled sample were also determined to be significantly different, however there was no difference among the 60, 90, and 120 min. samples in any of the attributes. Bleeding at some point after the fish were placed on the ice did reduce redness scores, however some degree of redness was retained the longer the time to initiate bleeding. The time interval before bleeding did not affect the sensory or color meter scores. In the raw filet (Figure 2), as with the whole fish, a significant difference was seen between the 0 time bleed and the no bleed samples in appearance, redness, and vein prevalence. Odor was not rated as distinguishable among any of the samples. Cooking the filet (Figure 3) eliminated any significant redness among any of the samples, however the no bleed, 60, 90, and 120 min. groups were significantly (p<0.05) darker in appearance than the 0 time bleed sample. Evidently this was due to the blood coagulating and providing a brown cast to the filet. Preference seemed to be less affected in the cooked sample indicating that the presence of redness or blood can have a detrimental affect on panelists' opinion of the product. As before, there were no differences in odor among the cooked samples. Texture or taste (Figure 4) was not significantly affected by any of the treatment groups. Due to the size of the sample portion, panelists were unable to be visually influenced by the bleed times and rated all of the samples equally. Results of the color meter analysis are presented in Table 3. The data showed that whole not bled fish had a higher darker color (L* value) and redness score (a* value) than the 0 time bled fish. The values for 60, 90, and 120 min. were not different significantly and were between the not bled and 0 time bled fish. Raw not bled filets also showed a significantly higher red component when compared to the fish bled at 0 time. Redness score also increased with increasing time to bleed. As was the case with the sensory data, the chroma meter results provided more evidence that cooking can reduce or eliminate the redness in the filet. The a* value was more negative for the cooked versus the raw filet. Overall, the chroma meter was an excellent analytical tool that provided an objective, numerical value which could be used to predict panelists scores.

Summary

These studies present numerous techniques that may be included as part of the operational procedures and may or may not affect the overall quality of an aquaculturally produced product. The killing method study indicated that all of the commonly used sacrificial methods had no adverse effect on texture. However, the use of carbon dioxide may be the method of choice since it is rapid, utilizes low cost technology, and presents a minimum health hazard to workers when compared to the other slaughter techniques. A rapid method of killing is suggested since the textural analysis revealed that stress will toughen filet meat. In addition the processing of filets post-rigor rather than pre-rigor resulted in a higher percent yield of as much as 5 percent. The bleed study indicated that bleeding the fish only affects the visual appearance of the whole fish, raw filet, and cooked filet. Bleeding did not affect odor, texture, or percent yield, however non
bled fish did seem to have an affect on the presumed quality level of the product as indicated by panelist preference scores. Bled fish seemed to impart a lighter appearance to the raw product which most consumers relate to better quality and would accordingly provide a positive influence at the point-of-sale.

Table 1. Percent Yield Data for Various Killing Methods.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Bled</th>
<th>Unbled</th>
<th>Bled Pre&amp;Pos</th>
<th>Unbled Pre&amp;Pos</th>
<th>All Trmts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control stressed</td>
<td>3.6</td>
<td>1.6</td>
<td>39.2</td>
<td>42.1</td>
<td>40.6</td>
</tr>
<tr>
<td>Control non-stressed</td>
<td>-1.3</td>
<td>2.1</td>
<td>39.5</td>
<td>40.0</td>
<td>39.8</td>
</tr>
<tr>
<td>CO₂ stressed</td>
<td>5.3</td>
<td>1.7</td>
<td>39.8</td>
<td>38.5</td>
<td>39.2</td>
</tr>
<tr>
<td>CO₂ non-stressed</td>
<td>1.3</td>
<td>4.0</td>
<td>38.6</td>
<td>39.4</td>
<td>39.0</td>
</tr>
<tr>
<td>Electroshock stressed</td>
<td>1.0</td>
<td>5.6</td>
<td>40.4</td>
<td>41.7</td>
<td>41.0</td>
</tr>
<tr>
<td>Electroshock non-stressed</td>
<td>3.5</td>
<td>-1.6</td>
<td>38.8</td>
<td>37.4</td>
<td>38.2</td>
</tr>
<tr>
<td>MS222 stressed</td>
<td>0.9</td>
<td>3.4</td>
<td>38.3</td>
<td>38.9</td>
<td>38.6</td>
</tr>
<tr>
<td>MS222 non-stressed</td>
<td>5.3</td>
<td>5.5</td>
<td>39.5</td>
<td>38.8</td>
<td>39.1</td>
</tr>
<tr>
<td>Slow chilling stressed</td>
<td>3.2</td>
<td>3.0</td>
<td>37.9</td>
<td>38.1</td>
<td>38.0</td>
</tr>
<tr>
<td>Slow chilling non-stressed</td>
<td>5.2</td>
<td>1.7</td>
<td>39.1</td>
<td>39.1</td>
<td>39.7</td>
</tr>
<tr>
<td>Fast chilling stressed</td>
<td>-1.6</td>
<td>0.2</td>
<td>37.6</td>
<td>37.8</td>
<td>37.7</td>
</tr>
</tbody>
</table>

Bled and unbled data = post-rigor yield minus pre-rigor yield.

Table 2. Proximate analysis data for various killing methods.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Protein</th>
<th>ADF</th>
<th>Lipids</th>
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</thead>
<tbody>
<tr>
<td>Control stressed</td>
<td>82.4</td>
<td>0.0</td>
<td>10.9</td>
</tr>
<tr>
<td>Control non-stressed</td>
<td>89.6</td>
<td>2.0</td>
<td>4.9</td>
</tr>
<tr>
<td>Electroshock stressed</td>
<td>86.8</td>
<td>0.0</td>
<td>6.1</td>
</tr>
<tr>
<td>Electroshock non-stressed</td>
<td>87.2</td>
<td>1.6</td>
<td>7.8</td>
</tr>
<tr>
<td>CO₂ stressed</td>
<td>79.2</td>
<td>0.0</td>
<td>14.5</td>
</tr>
<tr>
<td>CO₂ non-stressed</td>
<td>91.2</td>
<td>0.0</td>
<td>3.0</td>
</tr>
<tr>
<td>MS222 stressed</td>
<td>91.2</td>
<td>0.4</td>
<td>5.3</td>
</tr>
<tr>
<td>MS222 non-stressed</td>
<td>89.6</td>
<td>2.6</td>
<td>7.8</td>
</tr>
<tr>
<td>Slow chilling stressed</td>
<td>77.6</td>
<td>2.0</td>
<td>16.7</td>
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<tr>
<td>Slow chilling non-stressed</td>
<td>74.4</td>
<td>0.2</td>
<td>16.1</td>
</tr>
<tr>
<td>Fast chilling stressed</td>
<td>78.8</td>
<td>1.4</td>
<td>17.4</td>
</tr>
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</table>
Table 3. Color Analysis Bleeding Study.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>L</th>
<th>a</th>
<th>b</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Bleed Whole</td>
<td>70.79</td>
<td>5.57</td>
<td>10.44</td>
</tr>
<tr>
<td>Raw Filet</td>
<td>43.42</td>
<td>2.93</td>
<td>-1.01</td>
</tr>
<tr>
<td>Cooked Filet</td>
<td>77.82</td>
<td>0.10</td>
<td>12.76</td>
</tr>
<tr>
<td>0 min Time Whole</td>
<td>79.05</td>
<td>-2.56</td>
<td>10.20</td>
</tr>
<tr>
<td>Raw Filet</td>
<td>45.35</td>
<td>0.12</td>
<td>-2.72</td>
</tr>
<tr>
<td>Cooked Filet</td>
<td>82.11</td>
<td>-2.21</td>
<td>9.83</td>
</tr>
<tr>
<td>60 min Time Whole</td>
<td>74.41</td>
<td>3.36</td>
<td>10.62</td>
</tr>
<tr>
<td>Raw Filet</td>
<td>43.92</td>
<td>1.48</td>
<td>-1.59</td>
</tr>
<tr>
<td>Cooked Filet</td>
<td>81.22</td>
<td>-0.49</td>
<td>11.22</td>
</tr>
<tr>
<td>90 min Time Whole</td>
<td>71.42</td>
<td>5.34</td>
<td>10.40</td>
</tr>
<tr>
<td>Raw Filet</td>
<td>45.88</td>
<td>0.84</td>
<td>-2.32</td>
</tr>
<tr>
<td>Cooked Filet</td>
<td>78.43</td>
<td>-0.40</td>
<td>11.67</td>
</tr>
<tr>
<td>120 min Time Whole</td>
<td>72.21</td>
<td>5.28</td>
<td>9.50</td>
</tr>
<tr>
<td>Raw Filet</td>
<td>42.74</td>
<td>1.93</td>
<td>-1.82</td>
</tr>
<tr>
<td>Cooked Filet</td>
<td>78.53</td>
<td>-1.06</td>
<td>11.99</td>
</tr>
</tbody>
</table>
Figure 1. Whole fish sensory scores versus bleed times.

Figure 2. Raw filet sensory scores versus bleed times.
Figure 3. Cooked filet sensory scores versus bleed times.

Figure 4. Cooked taste sensory scores versus bleed times.
Hybrid Striped Bass Recirculating Facility, Number 1

The effects of storage temperature on microbiological populations of hybrid striped bass were studied. Filets were obtained from both bled and unbled hybrid striped bass and stored for up to 21 days at 29°F (-1.7°C), 32°F (0°C), and 36°F (2°C). Samples were withdrawn on day 3, 5, 8, 10, 13, 15, 17, and 21. The analysis started on the third day after slaughter. This time period was selected to coincide with the time frame in which processed fish are placed in both the distribution channel and retail markets.

Five swabs, each of one square inch, were taken from each fish and placed in 5 ml of peptone buffer. Samples were spread plated onto Standard Methods Agar and incubated at 35°C for 48 hours.

Surface swab counts showed an increase in microbial populations over time. Microbial counts increased fastest at the 36°F (2°C) storage temperature followed by the 32°F (0°C), and then the 29°F (-1.7°C). Figures 8 through 10 show the microbiological data for univiscerated, eviscerated, and fileted fish.

Samples were also taken for a sensory analysis at the same time the microbiological test were performed. The panelists were requested to state their preferences in three characteristics in the raw fish:

1. appearance,
2. odor, and
3. visual texture

and four characteristics in the cooked fish:

1. appearance,
2. odor
3. taste, and
4. texture.
The following 9 point hedonic scale recommended by the National Marine Fisheries Service, U.S. Department of Commerce, was used:

- 9 = excellent
- 8 = very good
- 7 = good
- 6 = fair
- 5 = borderline
- 4 = slightly poor
- 3 = poor
- 2 = very poor
- 1 = inedible

A reference sample (frozen the day of slaughter) was used for a comparison.

Fish stored 36°F (2°C) were deemed to be poor (or below) (a sensory score below 3) after 13 days for all characteristics.

Sensory analysis of the filets cut from unbled fish and held at 32°F (0°C) was terminated after 15 days since the taste panelists judged the samples as spoiled.

Sensory analysis of the fillets cut from bled fish and stored at 32°F (0°C) was discontinued after 17 days.

Sensory analysis of fish stored at 29°F (-1.7°C) was stopped after 21 days.

The results showed that shelf life is dependent on storage temperature and that the lower the storage temperature the longer the shelf life. A small difference in storage temperature can result in a significant increase in shelf life. The primary reason for the extended shelf life is to reduce spoilage of micro flora on the fish.

No major differences were noticed by the panelists between bled and unbled fish in any characteristic other than the limited shelf life extension. The economic and consumer acceptability aspects of this shelf life extension cannot be properly evaluated at this time. It is possible that bleeding can have a significant effect on textural properties and frozen shelf life. At this time, bleeding immediately after killing is recommended.

Figures 1 through 7 show the trends for raw and cooked sensory characteristics.

**Hybrid Striped Bass Recirculating Facility, Number 2**

A Study was undertaken to identify human pathogens that may be capable of growing in hybrid striped bass recirculating facilities. Since some of the bass introduced into the recirculating systems were initially grown in estuarine water ponds, it is possible that pathogenic microorganisms of marine origin may be transferred with the fish.
One hundred milliliters of raceway water was filtered through a 45 micron filter and placed in Selenite Cysteine Broth (Difco, Inc.). After 24 hours the samples were then streaked onto XLD and Bismuth Sulfite Agar (BSA) and incubated at 37°C for 48 hours. Colonies were screened using the API Rapid SYS to detect Salmonella colonies. No colonies were Salmonella positive.

Cultures received from Facility Number 2 were streaked on TCBS agar plates, T-soy agar plates, and T-soy agar plates containing 2.5% sodium chloride. Cultures were also tested for oxidase reaction and Gram-stained. Presumptive positive colonies were then stabbed into motility agar and TSI (Triple Sugar Iron) agar tubes. All cultures were then biochemically profiled using the API Rapid NFT (Analytab Products, France).

Culture identification number 221101 (Unknown origin), 241101 (isolated from kidney), 241102 (from kidney), and 241105 (isolated from spleen) grew visually same colony types on T-soy agar, T-soy agar + 2.5% NaCl, and TCBS agar. All four were identified as Vibrio vulnificus by gram stain morphology, motility, oxidase reaction, TSI (Triple Sugar Iron) agar slant reaction, and Rapid NFT API Test for non-fermentative, gram-negative bacteria. This organism is an opportunistic pathogen that needs some underlying stress or disease before invasion. However, these fish already were stressed by other disease and salt content of water. Given these conditions, this organism can be quite detrimental to the fish population. Added salt that is used to control fungi in the tanks will provide an excellent environment for the Vibrio spp. Most of the species in this genus need at least 2.5% salt to grow. If adding salt for antifungal purposes, we suggest adding the salt to allow for antifungal action then decrease the salt to eliminate the overgrowth of Vibrio as soon as possible.

Culture identification number 221103 (unknown origin) was originally thought to be a Vibrio spp. after showing motility and being oxidase positive. However, the organism did not Gram stain a uniform Vibroid shape. The stain showed short gram-negative rods. Furthermore, the API test could not conclusively identify the organism as a Vibrio. The test actually showed it may be a Pseudomonas putrefaciens. The organism also did not grow on TCBS, eliminating many species of Vibrio. TSI reaction finally eliminated the possibility the organism was Vibroid, since hydrogen sulfide gas was produced. This reaction further upheld that this organism was Pseudomonas putrefaciens. This isolate, unlike Vibrio vulnificus, is not a pathogen but a spoilage organism found in soil and water. This organism is not a cause of lesions or illness.

Culture number 221105 was identified as Vibrio cholera. Further biochemical test showed that this may be V. cholera. However, serological testing would need to be done to conclusively identify this organism. Our laboratory did not perform the test.

Finally, number 241109 was identified by API and other biochemical tests as Vibrio fluvialis with 99.4% accuracy. This organism can be a human pathogen causing diarrhea. It was not known if it is a fish pathogen.

Some fish grown in the recirculating facility were judged to have a muddy earthy flavor which resulted in their being rejected by some dealers. The fish had a sensory profile similar to catfish.
containing geosmin or MIB (methylisoborneol). In order to determine the cause of the unacceptable order or flavor, raceway water samples were extracted in vegetable oil and run on a Hewlett Packard 5980 gas chromatograph using the method described by Dupuy.

MIB was detected in all samples tested while geosmin was not detected. A microbiological study of the rotating biological contactor (RBC) revealed the presence of Streptomyces. This organism is a known producer of MIB and appeared to be the cause of the off flavor.

Figures 11 through 13 show the chromatograms for these samples.

**Trout Recirculating Facility**

A study of microbiological pathogens was conducted on various rainbow trout production systems using concrete and mud raceways, silos, and a recirculating system.

Of the pathogens isolated in this study, none were present at levels sufficient to pose a risk to consumers who will be cooking their own products. This, however is not the case with processors who will be creating ready to eat products. *Clostridium botulinum* has been shown in the past to be a problem in the smoked fish industry. Care needs to be taken to ensure that the conditions are present to adequately inhibit the outgrowth of toxin in both aerobically and modified atmospheric packaging (proper water phase-salt and storage temperature). A proper processing temperature for the ready-to-eat product also needs to be reached that would destroy any *Listeria monocytogenes* present, and secondly care needs to be taken to ensure that cross contamination of processed and ray product does not occur.

Pathogen levels and the general microbial quality remained unchanged with respect to season.

There was also no evidence that would support the safety of one aquaculture system over another. This fact may prove useful in regions of the United States where culture of rainbow trout is severely limited by the lack of water sources that supply the high flow rate needed to raise trout.

Results of the study are presented in Table 1 through 5.

Fish muscle samples from one recirculating facility had a musty-earthy flavor characteristic of geosmin or MIB. A gas chromatographic analysis indicated the presence of geosmin. A compound was produced by algae growing in a pond in which the fish were placed for a short period of time prior to harvest. Consequently, stream or pond water may become contaminated with geosmin and the fish will be required to proceed through a deputation procedure prior to sale.
Table 1. Plate Counts (Mesopholic and Psychotrophic) for Whole Rainbow Trout and Fillets (CFU/g: Log transformation)

<table>
<thead>
<tr>
<th>Plant</th>
<th>Whole Fish</th>
<th>Fillets</th>
<th></th>
<th></th>
<th></th>
<th></th>
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<th></th>
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<tbody>
<tr>
<td>1</td>
<td>1.74 x 10^4</td>
<td>2.95 x 10^4</td>
<td>2.29 x 10^4</td>
<td>3.09 x 10^4</td>
<td>4.68 x 10^4</td>
<td>5.62 x 10^4</td>
<td>7.24 x 10^4</td>
<td>1.05 x 10^5</td>
</tr>
<tr>
<td>2</td>
<td>3.55 x 10^3</td>
<td>2.51 x 10^4</td>
<td>1.95 x 10^4</td>
<td>1.55 x 10^4</td>
<td>2.63 x 10^4</td>
<td>3.72 x 10^4</td>
<td>4.9 x 10^4</td>
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</tr>
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<td>3</td>
<td>2.04 x 10^3</td>
<td>7.76 x 10^4</td>
<td>5.37 x 10^4</td>
<td>1.1 x 10^4</td>
<td>2.95 x 10^4</td>
<td>3.8 x 10^4</td>
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<tr>
<td>4</td>
<td>1.15 x 10^3</td>
<td>1.48 x 10^5</td>
<td>1.95 x 10^4</td>
<td>3.02 x 10^4</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5**</td>
<td>2.00 x 10^4</td>
<td>7.41 x 10^4</td>
<td>5.01 x 10^4</td>
<td>7.24 x 10^4</td>
<td>-</td>
<td>-</td>
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<td>-</td>
</tr>
</tbody>
</table>

** indicates recirculating system
- firm does not have process operations

Table 2. Coliform, Fecal Coliform, and _Escherichia coli_ counts for rainbow trout rearing water and fillets (MPN/g o ml: Log transformation)

<table>
<thead>
<tr>
<th>Plant</th>
<th>Rearing Water (MPN/ml)</th>
<th>Fillets (MPN/g)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Col.</td>
<td>Fecal Col.</td>
<td><em>E. coli</em></td>
<td>Col.</td>
<td>Fecal Col.</td>
<td><em>E. coli</em></td>
<td>Col.</td>
<td>Fecal Col.</td>
</tr>
<tr>
<td>1</td>
<td>1.90</td>
<td>0.47</td>
<td>0.47</td>
<td>15.30</td>
<td>2.45</td>
<td>0.40</td>
<td>204.00</td>
<td>4.98</td>
</tr>
<tr>
<td>2</td>
<td>22.66</td>
<td>0.69</td>
<td>0.58</td>
<td>4.02</td>
<td>0.26</td>
<td>0.00</td>
<td>583.20</td>
<td>0.07</td>
</tr>
<tr>
<td>3</td>
<td>21.86</td>
<td>2.62</td>
<td>1.82</td>
<td>3.50</td>
<td>0.00</td>
<td>0.00</td>
<td>99.00</td>
<td>0.00</td>
</tr>
<tr>
<td>4</td>
<td>188.00</td>
<td>41.60</td>
<td>25.92</td>
<td>39.50</td>
<td>0.00</td>
<td>0.00</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5**</td>
<td>1.54</td>
<td>0.00</td>
<td>0.00</td>
<td>15.28</td>
<td>0.00</td>
<td>0.00</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

** indicates recirculating system
- firm did not have processing operations
ND – Not Detected
**Table 3.** *Listeria* Incidence and Quantitative Levels on Whole and Filleted Rainbow Trout in Late Summer (1994)

<table>
<thead>
<tr>
<th></th>
<th>Whole fish</th>
<th>Fillet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Plant 1</td>
<td>Plant 2</td>
</tr>
<tr>
<td>Number of Samples Positive for <em>Listeria spp.</em> (20 samples)</td>
<td>18 (90%)</td>
<td>10 (50%)</td>
</tr>
<tr>
<td>Average Counts for Positive Samples (MPN/g) Log Trans.</td>
<td>4.83</td>
<td>1.02</td>
</tr>
<tr>
<td>Samples positive for <em>Listeria monocytogenes</em></td>
<td>18 (90%)</td>
<td>7 (35%)</td>
</tr>
<tr>
<td>Other <em>Listeria spp.</em> Isolated</td>
<td>none</td>
<td><em>L. innocua</em> 5 (25%)</td>
</tr>
</tbody>
</table>

** indicates recirculating system

**Table 4.** Data for *Listeria* Incidence and Quantitative Levels on Whole and Filleted Rainbow Trout in Late Spring (1995)

<table>
<thead>
<tr>
<th></th>
<th>Whole fish</th>
<th>Fillet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Plant 1</td>
<td>Plant 2</td>
</tr>
<tr>
<td>Number of Samples Positive for <em>Listeria spp.</em> (20 samples)</td>
<td>14 (70%)</td>
<td>5 (25%)</td>
</tr>
<tr>
<td>Average Counts for Positive Samples (MPN/g) Log Trans.</td>
<td>1.83</td>
<td>1.27</td>
</tr>
<tr>
<td>Samples positive for <em>Listeria monocytogenes</em></td>
<td>9 (45%)</td>
<td>4 (20%)</td>
</tr>
<tr>
<td>Other <em>Listeria spp.</em> Isolated</td>
<td><em>L. innocua</em> 10 (50%)</td>
<td><em>L. innocua</em> 2 (10%)</td>
</tr>
</tbody>
</table>

** indicates recirculating system
Table 5. *Clostridium botulinum* Incidence and Quantitative Levels on Whole Rainbow Trout in Late Summer (1994) and Late Spring (1995)

<table>
<thead>
<tr>
<th>Number of Samples Positive for presumptive <em>C. botulinum</em> spores</th>
<th>Late Summer (1994)</th>
<th>Plant 1</th>
<th>Plant 3</th>
<th>Plant 4</th>
<th>Plant 2</th>
<th>Plant 5**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Late Spring (1995)</td>
<td>13 (65%)</td>
<td>14 (70%)</td>
<td>16 (80%)</td>
<td>9 (45%)</td>
<td>19 (95%)</td>
<td></td>
</tr>
<tr>
<td>Average Counts for Positive Samples (MPN/G) Log trans.</td>
<td>Late Summer (1994)</td>
<td>1.60</td>
<td>0.46</td>
<td>0.54</td>
<td>1.76</td>
<td>1.69</td>
</tr>
<tr>
<td></td>
<td>Late Spring (1995)</td>
<td>0.68</td>
<td>0.89</td>
<td>2.33</td>
<td>4.31</td>
<td>1.40</td>
</tr>
</tbody>
</table>

** indicates recirculating system
Figure 1. Raw Striped Bass Appearance

[Graph showing sensory score over days for different temperature and bleeding conditions]
Figure 2. Raw Striped Bass Odor
Figure 3. Raw Striped Bass Texture
Figure 4. Cooked Striped Bass Appearance
Figure 5. Cooked Striped Bass Odor

Sensory Score vs. Day

- 29 F Bled
- 29 F Unbled
- 32 F Bled
- 32 F Unbled
- 36 F Bled
- 36 F Unbled
Figure 6. Cooked Striped Bass Taste
Figure 7. Cooked Striped Bass Texture
Figure 8. Surface Swab Uneviscerated Striped Bass
Figure 9. Surface Swab Eviscerated Striped Bass

Log CFU/sq.in.

Day

29 F Bled
29 F Unbled
32 F Bled
32 F Unbled
36 F Bled
36 F Unbled
Figure 10. Surface Swab Striped Bass Filets

Log Cfu/sq.in.

Day

0  5  10  15  20  25
0  2  4  6  8 10

- 29 F Bled
- 29 F Unbled
* 32 F Bled
* 32 F Unbled
* 36 F Bled
* 36 F Unbled
Figure 11. Sample A: Before Ozonation

MIS

GEOSMIN
Figure 12. Sample B: After Ozonation

MIS

GEOSMIN
Figure 13. Sample C: Raceway 1A

MIS

GEOSMIN
Utilization of Soft-Shell Crawfish and Alligators Grown in Recirculating Aquaculture Systems

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Louisiana Cooperative Extension Service
Louisiana State University Agricultural Center

Introduction

Alligators and soft-shell crawfish are commercially important aquaculture species. Many of the challenges associated with growing these animals in aquaculture and recirculating systems have been researched and data published (Culley et al., 1989; Culley et al., 1985; Culley et al., 1985a; Joanen and McNease, 1979). One major area of concern for commercial production is post-harvest handling. Problems inherent to food and product safety and quality must be addressed to facilitate successful introduction into consumer markets and to ensure customer satisfaction. The purpose of this paper is to present information pertaining to the use and post-harvest handling procedures of alligators and soft-shell crawfish.

Soft-Shell Crawfish Production and Processing

Hard-shell crawfish are a traditionally and commercially important crustacean seafood product in Louisiana. The two most significant commercially available species are the Red Swamp crawfish (Procambarus clarkii) and the White River Crawfish (Procambarus zonangulus). Although both species may be found in the same growing areas and are mingled during harvest activities, the Red Swamp crawfish has many favorable characteristics associated with soft-shell crawfish production. The production of softshell crawfish is seasonal and mirrors that of hard crawfish production since hard crawfish must be used to stock soft crawfish shedding trays. The peak of production is in the late winter, throughout the spring and early summer. Crawfish have been successfully raised in pond culture for decades. In 1995, there were more than 60,000 acres in production in the state. An estimated 45,000,000 pounds of live crawfish were sold into commerce from pond production. The preponderance of crawfish produced from ponds is sold as one of three products: (1) live, whole (2) cooked, seasoned and whole, or as (3) cooked, peeled and deveined meat. Soft-shell crawfish are another commercially important form of crawfish aquaculture. Although soft-shell crawfish have been locally consumed, commercial production was not possible until the emergence and development of intensive culture systems in 1985 (Culley et al., 1985).

Unlike other crawfish products, where only the peeled and deveined tail meat is eaten, soft-shell crawfish are basically eaten whole, with only minor butchering required. The production of soft-shell crawfish is dependent upon natural cycles of growth and maturation. As part of these life cycles, crawfish periodically shed this hard exoskeleton and form new and larger ones.
Immediately after the shedding process, the exoskeleton is soft, pliable and edible along with the meat and organs of the animal. The exoskeleton must remain soft throughout the post-harvest period for optimal quality.

Post-harvest processing of soft-shell crawfish is relatively simple but may vary considerably from producer to producer. In general, the aquaculture producer also conducts the processing procedures. Prior to molting, crawfish extract calcium from the old exoskeleton and store it in the form of calcium stones in the head. These hard stones are called gastrolith. For obvious reasons of consumer safety, these stones must be removed before packaging. Removal requires cutting off the face of the crawfish and extracting the stones by hand. In some cases, this may be the only processing step the producer chooses to conduct. In other cases, the producer may choose to remove all internal organs and the carapace. The latter butchering procedure is considered standard for the industry. After butchering, the crawfish are packaged. The resulting product may be presented to users as either a fresh or frozen product. The industry standard is to package and freeze the product.

**Quality Parameters for Soft-Shell Crawfish**

Seafoods are among the most perishable foods. Proper packaging may minimize some of the negative changes that occur in seafoods. Commonly identified detrimental changes in seafoods include oxidative rancidity, dehydration, drip loss, color and flavor changes and textural changes. Soft-shell crawfish are highly sensitive to processing and preservation methods and are, impart, subject to all of the above detrimental changes. In addition, improper post-molting handling may result in undesirable product offered to market. Since soft-shell crawfish are processed while alive and raw and are frozen almost immediately after butchering, microbiological considerations are not considered significant. However, texture, drip loss and factors associated with freezing have been identified as parameters that affect the overall quality of soft-shell crawfish.

**Exoskeleton Texture**

Production of soft-shell crawfish in recirculating systems begins with the introduction of hard-shell crawfish to the shedding system. Crawfish molt unpredictably but generally during daylight hours (Culley and Duobinis-Gray, 1989). If crawfish are not removed quickly after molting, the new exoskeleton begins to harden and renders the crawfish unacceptable. To prevent this, freshly molted crawfish should be quickly removed from the system molting trays and placed under refrigeration. Studies have shown that temperature reduction retards the onset of exoskeleton hardening (Huner et al., 1978). Park et al., 1989, recommended removal of freshly molted crawfish and placing them in refrigerated water at 2°C until packaging. These same researchers also recommended that soft-shell crawfish be packaged the same day they are removed from molting trays. Crawfish left overnight before packaging, even in refrigerated water, may exhibit shell hardening to the point of being unacceptable. In summary, producers must periodically check molting trays throughout the daylight hours and quickly remove molted crawfish and place them in refrigerated water (2°F). All crawfish must be prepared and packaged the same day as they molt for highest quality.
Culley, (1989b), noted that commensal organisms are associated with freshly molted soft shell crawfish. Although harmless, these organisms detract from the overall appearance of the product. He found that a refrigerated holding solution of 1% NaCl was effective in removing the organisms without harm to the product. Park, (1990) evaluated the effectiveness of using chlorinated holding solutions on the overall quality of soft-shell crawfish. Chlorinated holding solutions not only remove the commensal organisms but also anesthetize or kill the crawfish, allowing for easier packaging. The researcher experimented with placing crawfish in a holding solution containing 2.6 ppm sodium hypochlorite solution for 8 hours or by dipping for 15 minutes in a 2.6 ppm sodium hypochlorite solution and holding in a water solution for 8 hours. A taste panel evaluated the resulting product. The taste panel judged the chlorinated treated crawfish to be less juicy and tougher in texture when compared to a control of water solution. However, it was determined that placing the crawfish in the chlorine solution for up to 8 hours created no serious deterioration of quality. The researcher recommended the 15-minute chlorine dip as satisfactory. Although the researcher did not conduct any microbiological tests, he projected a reduction in microbiological numbers based on similar dips with other seafoods.

Freezing Method

The production of soft-shell crawfish is seasonal. Hard-shell crawfish must be available for stocking molting trays. Traditional hard-shell crawfish are readily available for this purpose from November until the end of May. Variability of the season may provide crawfish earlier or later than these dates. Even within these predictable seasonal parameters, the quantity of available quality crawfish may not be dependable. Since the hard crawfish are only seasonally available, soft-shell crawfish are seasonal. To provide crawfish for year round consumption and to stabilize market demand, soft-shell crawfish are generally packaged and frozen. Freezing any seafood product can have detrimental consequences to the final product. Of particular concern are texture changes, oxidative rancidity, moisture loss and flavor changes. Park et al., (1990), clearly demonstrated that fresh, unfrozen soft-shell crawfish were more desirable than frozen ones. Of particular interest is the impact the digestive organ in the cephalothorax called the hepatopancreas has on frozen crawfish. Commonly called “fat,” this organ is generally consumed and is considered highly favorable by most consumers familiar with crawfish. Unfortunately, the tissue from this organ has been associated with textural changes and off flavors as a result of frozen storage. Proteolytic enzymes in the hepatopancreas are believed to be responsible for the mushiness in some crawfish products. Generally, this mushiness is attributed to crawfish products that were processed using raw crawfish meat or undercooked or blanched crawfish meat. Thoroughly cooked crawfish do not exhibit mushiness since a sufficient heat treatment will inactivate the proteolytic enzymes. Since soft-shell crawfish are processed and stored raw, product texture during freezing and during fresh or frozen storage is a major concern. The hepatopancreas is also thought to be responsible for rancidity, resulting in an undesirable off flavor that develops in frozen crawfish products. Other factors of concern during freezing are textural and flavor changes caused by the freezing process. Mechanical damage may occur as a result of ice crystal formation. It is a general premise that the faster the product is frozen, the smaller the ice crystals will be and the less damage to cellular material. This type of damage
would result in a toughening in texture and an increased drip loss. For this reason, many food manufacturers have turned to cryogenic freezing.

Park (1990) evaluated the effects of mechanical freezing (-20°C) compared to cryogenic freezing (-100°C) on packaged soft-shell crawfish. Interestingly enough, he was not able to demonstrate any advantage to cryogenic freezing to mechanical freezing using his parameters. In fact, his data demonstrated greater textural damage and drip loss caused by cryogenic freezing than mechanical freezing.

**Drip Loss**

Drip loss is always a concern for seafood processors. Although the moisture lost through desiccation or through weeping of the tissues was at one time part of the edible product, these losses must be taken into account when declaring net weight and profit. Excessive drip loss can result in inferior product quality. Processors make every effort to minimize these losses. Drip loss may be affected by freezing methods as already discussed. There are many other factors that influence the drip loss in soft-shell crawfish, including cold storage temperature variables (multiple freeze/thaw cycles), packaging methods and thawing procedures. One major variable that affects drip loss in soft-shell crawfish is butchering procedures. The industry standards for butchering requires complete removal of the carapace and all internal organs. The typical yield from this procedure is only about 50% of the original weight (Park, 1990). Although much of the weight loss can be attributed to parts removal, other weight loss is caused by drip loss and can be attributed to drip loss of body fluids during butchering, exposure of unprotective tissues to desiccation and dehydration during freezing and freezer storage, and drip loss during thawing and product preparation.

**Grading**

Soft-shell crawfish come in various sizes depending on the size of the hard crawfish supplied to the producer. Unlike other crustaceans such as shrimp that are sized according to count (that is, the number of shrimp per pound), soft-shell crawfish are graded according to length. When the industry was emerging in the 1980s, there were various attempts to standardize a grading system. The one system most accepted by the industry today was developed by the Louisiana Seafood Promotion and Marketing Board. The grades are:

- Jumbo.................4 inches and over
- Extra Large............3.5-4 inches
- Large..................3-3.5 inches
- Medium.................3 inches and under

It is interesting to note there is no small size designation. The Board points out that the most common sizes in the industry are Extra Large and Large.
Packaging

Packaging of soft-shell crawfish was identified by the industry in the late 1980s as one of the major concerns for product presentation. Prior to that time, the product was placed in self sealing polyethylene bags with water, followed by mechanically freezing. This was not an appealing or attractive package to users and it was difficult to store and thaw the product. The addition of the water as a packaging medium added additional expense for freezing and unwanted weight during storage. For the most part, users could not adequately examine the individual crawfish for defects and since all the crawfish in the package were frozen in block form, portioning was difficult. The polyethylene bags required manipulation to flatten the bags just before freezing. Such handling severely damaged the product.

Park (1990) evaluated the effects of using more attractive packaging materials for softshell crawfish. He compared crawfish packaged in water in a zip pouch polyethylene bag to a shrink wrap in a Styrofoam dish. Interestingly, he found that the zip pouch package had significantly less drip loss of moisture than the more attractive tray package. Although the zip pouch had a reasonable drip loss as low as 5%, the tray package had a drip loss as high as 15%. Even though the study was conducted over a 36-week period, drip loss data were consistent throughout the study. There are many reasons for these results. Although the tray package was considerably more attractive than the zip pouch, the material did not provide a sufficient barrier to the product. Consequently, desiccation may have occurred. The product in the pouch was essentially protected from dehydration by the surrounding glaze. Park, 1990, also found that soft-shell crawfish packages in the zip pouches had significantly greater flavor intensity. In addition, when thawed and removed from the package, the product in the zip pouches had a superior physical appearance. Although this study clearly indicated that the presentation and utility of the product could be significantly improved by using the tray pack product, this particular product form did not improve the organoleptic qualities of the product. Today, with some improvements, the industry is currently using a tray pack with an overwrap. Each tray contains two dozen individually packaged crawfish. To absorb excess moisture (called “ink” because of the dark color) released during packaging, freezing and thawing, an absorbent pad is placed in the bottom of the tray.

Summary

The soft-shell crawfish industry has had many quality challenges since its commercial development. Today, the industry has developed practices that have gained acceptance and are considered standard operations. These include butchering, grading and packaging.

Alligator Production

The American alligator (Alligator mississippiensis) is an aquatic reptile found in subtropical southeastern United States. These animals are particularly abundant in Louisiana and Florida. At one point in the 1960s, the American alligator was classified as endangered because of overhunting and poaching. With effective protective measures in place, population level grew to
sufficient numbers that allowed a controlled harvest program that began in 1972 and continues today (Moody, 1980). For management purposes, the wild hunting season is conducted for approximately 30 days and is conducted primarily in September. In 1995, more than 24,600 alligators were harvested from the wild in Louisiana. Alligators are also a species that has adapted well to aquaculture. In 1994, more than 135,000 alligators from aquaculture were reared and slaughtered.

There are physical and quality differences between alligators harvested from the wild and those from aquaculture. One of the most obvious is size. Alligators harvested from the wild may be quite variable in length, ranging from 4 feet to more than 13 feet. The average wild captured alligator is approximately 7 feet. On the other hand, aquaculture grown alligators are consistent in size. Most are harvested when 3 to 4 feet in length. Wild caught alligators may be many years old, but most aquaculture-grown alligators are less than 2 years old. This could affect the taste and other quality factors in the meat. Because wild alligators must be captured and slaughtered based on a management plan (30 days as mentioned previously), the bulk of this source is harvested in a very short interval. This creates a logistical problem in processing and handling thousands of pounds of meat brought onto the market in a matter of a few weeks. Conversely, aquaculture alligators may be slaughtered at the decretion of the producer at any time.

**Slaughter Operations**

Alligators grown in aquaculture are selectively harvested and humanely slaughtered under controlled conditions. Slaughter facilities are designed to meet state processing requirements. Long tables at a comfortable working height are used to process the animals. Alligators are manually skinned by highly skilled skinners using a sharp knife. It is important that the animals be skinned soon after killing. The skins are scraped to remove traces of muscle or fat tissue, rinsed in clean water and then salted. Salt should be generously applied and rubbed into all parts of the skin in a layer 1/2 to 1 inch thick (Louisiana Fur and Alligator Advisory Council). Salt will remove excess water and will retard bacterial growth. The Council further recommends that after salting, the skins should be rolled into a tight ball for several days to drain excess water. After 3-5 days the skins should be unrolled and resalted. The remaining carcass can be cut up for meat and offered for sale.

**Alligator Hide Quality and Processing**

The alligator industry is based on the use of the skin or hide. The value of the skin is based on many factors including the overall length and belly width of the animal, the quality or condition of the hide as a result of processing and storage and the prevailing market price at the time of sale. The demand for certain types of alligator hides affects markets significantly. For example, there are two types of hides, belly patterns and horn backed patterns. The belly pattern requires making cuts that leave the leg, sides and belly skin intact and in one piece. Hides with belly patterns are the most common and have the greatest demand for the “classic” leather market. Any alligator can be skinned out using the belly pattern. Horn backed patterned skins leave the leg, sides and backs intact. This cut is made by splitting the belly skin. This hide cut is not nearly as common as
the belly pattern and is used for making some types of boots, brief cases, etc. Horn backed pattern skins can be used only from alligators approximately 4 feet in length or less.

Depending on market conditions and demand, the belly width of the skin determines the overall value of the skin. Hide quality is of prime importance. Many factors establish the quality of alligator skins. An alligator is not easily skinned. Meat and fat pieces are difficult to remove and require careful hand scraping. Any meat or fat left on the skin significantly lowers the quality of the skin. The Louisiana Fur and Alligator Advisory Council advises that meat and fat left on the skin provide ideal conditions for bacteria to grow during the long storage period before tanning occurs.

Decomposition that begins in adhering meat pieces will ruin the skins. Two decomposition-related defects are called “red heat” and “slipping skins.” Fat will not permit the proper salt penetration and may leave greasy spots on the finished tanned hide. Another major defect that can occur during the skinning process is accidental skin cuts. Because the skin must be cut from the carcass with a sharp knife, there are many opportunities to nick or cut into valuable belly skin. Other skin defects such as “buttons” and scars may occur while the animal is still living and growing, and they affect the final quality. The American Classic Grading System is used to categorize cured hides at the time of sale.
Table 1. Alligator yield data

<table>
<thead>
<tr>
<th>Size</th>
<th>Length (In.)</th>
<th>Live wt.</th>
<th>Dressed wt., head off</th>
<th>Dressed wt., head off, after 12 hrs. @ 35°F</th>
<th>Skin wt.</th>
<th>Waste wt. (head, viscera and feet)</th>
<th>Cut of Meat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Tail</td>
</tr>
<tr>
<td>Small</td>
<td>56.5</td>
<td>18.1</td>
<td>11.5</td>
<td>11.0</td>
<td>2.9</td>
<td>4.1</td>
<td>3.8</td>
</tr>
<tr>
<td>Medium</td>
<td>77.5</td>
<td>49.4</td>
<td>30.5</td>
<td>30.4</td>
<td>7.9</td>
<td>11.1</td>
<td>10.2</td>
</tr>
<tr>
<td>Large</td>
<td>88.8</td>
<td>83.6</td>
<td>52.4</td>
<td>50.4</td>
<td>12.2</td>
<td>19.0</td>
<td>17.2</td>
</tr>
<tr>
<td>Extra Large</td>
<td>110.0</td>
<td>262.0</td>
<td>162.0</td>
<td>159.0</td>
<td>34.8</td>
<td>59.0</td>
<td>48.6</td>
</tr>
</tbody>
</table>

Source of data:
Table 2. Percentage yield of alligator on a live-weight basis

<table>
<thead>
<tr>
<th>Size</th>
<th>Dressed, head off</th>
<th>Waste (head, viscera, feet and skin)</th>
<th>Cut of Meat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Tail</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Leg</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Torso</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Jaw</td>
</tr>
<tr>
<td>Small</td>
<td>63.3</td>
<td>38.2</td>
<td>21.1</td>
</tr>
<tr>
<td>Medium</td>
<td>61.7</td>
<td>38.6</td>
<td>20.7</td>
</tr>
<tr>
<td>Large</td>
<td>62.6</td>
<td>37.3</td>
<td>20.6</td>
</tr>
<tr>
<td>Extra Large</td>
<td>61.8</td>
<td>36.0</td>
<td>18.5</td>
</tr>
</tbody>
</table>

Source of data:

Table 3. Proximal analysis of alligator meat

<table>
<thead>
<tr>
<th>Cut of Meat</th>
<th>Crude protein</th>
<th>Crude fat</th>
<th>Moisture</th>
<th>Ash</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>%</td>
<td>%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tail</td>
<td>21.3</td>
<td>1.5</td>
<td>76.5</td>
<td>1.3</td>
</tr>
<tr>
<td>Torso</td>
<td>21.1</td>
<td>1.2</td>
<td>73.9</td>
<td>1.3</td>
</tr>
<tr>
<td>Jaw</td>
<td>22.3</td>
<td>1.2</td>
<td>75.9</td>
<td>1.3</td>
</tr>
<tr>
<td>Leg</td>
<td>21.1</td>
<td>1.0</td>
<td>76.8</td>
<td>1.3</td>
</tr>
</tbody>
</table>

1 Analysis performed by LSU Feed and Fertilizer Laboratory.

Source of data:

Alligator Meat Processing

When alligator harvesting was approved in 1972, the primary commercial product was the valuable skin. At that time, most of the meat resulting from skin processing was discarded for several reasons. There were strict regulations on alligator parts that did not provide easy access to markets, and no commercial marketing channels were available. In the early 1980s, efforts to market the meat were begun. Moody, et al., did much of the pioneering work in use of alligator meat for intended human consumption. Processing regulations have been published by the Louisiana State Food and Drug. These regulations specify that all animal be butchered (including skinning) in facilities that meet all health and sanitation requirements for handling and processing human food.

Moody, et al., 1980, determined alligator meat yield data shown in Tables 1 and 2. For this study,
alligators were divided into four size categories. The data show that dressed weight and waste weight percentages were nearly proportional, the live weight, dressed weight, skin weight and waste weight increased more than that of length. Moody also made observations on the meat. Meat from the tail, torso and jaw is white to pink. The tail meat had characteristic internal and external bands of fat. All cuts of meat were extremely lean with no observed marbling. The leg meat was darker, with some fat deposits. Moody also conducted a proximal analysis of the meat and these data are in Table 3. It has been noted that fat imparts an off flavor in meat of wild harvested alligators and must be entirely removed before to cooking the meat. The off flavor is not noticeable in farm produced meat, probably due to diet and the young age. Observations indicate that alligator meat is stable under frozen condition.

Other parts of the alligator such as the teeth, head and feet are valued in the jewelry and novelty trade.

**Summary**

Alligator hides have been an important product of commerce for decades. The meat was viewed as a non-traditional dish until recent years when consumer demand established it as a stable and significant product of commerce. Quality factors for both the skin and the meat are important considerations for the producer.

**References**


Although a HACCP plan may not be required by law, developing such a plan for a commercial recirculating aquaculture facility makes sense for several reasons. A HACCP plan requires a thoughtful analysis of the production process, thus some potentially important management aspects may be reviewed and found to be of greater (or lesser) importance than previously thought. The HACCP process requires accurate record keeping. This will lead to better management if the present management strategy is weak in keeping records of various activities. Additionally, the HACCP development process may reveal that certain records are important and must be retained for a considerable length of time, while other records can be discarded after a brief time period. Finally, the HACCP process may facilitate discovering causes when things go wrong. It is inevitable in the aquaculture business that things will go awry. When this happens, insuring that the safety and wholesomeness of the product has not been compromised, discovering the root cause of the problem, and fixing the problem quickly will be foremost in the operators priorities. A good HACCP plan can help in all of these areas.

To begin the development process, a thorough and accurate process flow diagram must be developed. For the purposes of this presentation, a set of example figures, forms, and tables have been developed. These are for illustrative purposes only. A separate analysis will have to be conducted for each firm/operation that fits the specific situation. The examples used here, Small Fortune Aquaculture, is deemed to be a commercial scale (derives majority of income from this venture) recirculating system facility. For this firm, a hypothetical flow diagram (Figure 1.) was developed that included the following control points: water, feed, fish (incoming fingerlings), water treatment chemicals, therapeutics, the flora and fauna in the grow out unit, and “other” factors.

In this presentation, the control points water, feed, flora and fauna, and harvest will be discussed. The remaining points will be covered in the next presentation. For each control point, a determination must be made as to whether the point is critical. Each control point will be discussed separately.
Figure 1. Flow diagram for a commercial scale firm utilizing recirculating system technology.  Example – For Illustrative Purposes Only.

Water

This control point was determined to be critical. Possible defects include: water borne diseases, heavy metal contamination, pesticides, bacteria, industrial organics (PCBs), and PANO (Poly-Aromatic Nucleated Organics = petroleum decomposition products). The critical limits will be established based on the end use of the product (fish). Possible variables that would influence the critical limits include – food fish versus non-food fish (ornamentals), sensitivity of the fish being grown, and amount and degree of water usage. Preventative measures are limited to periodically analyzing the water by an independent laboratory with appropriate tests being conducted. The monitoring procedure includes twice annually analyzing the water and fish for appropriate hazards. If the water does not meet specifications: 1. Quarantine all fish exposed to this source, 2. Analyzed the fish for safety, and 3. Investigate the water source for source of the problem. Records for this critical control point should include: date the samples was taken, where the sample was taken, the person taking the sample, and the results of the analysis. Verification will include the facility manager reviewing the records after each sample period, confirming that the records and samples are accurate, and an annual review by the manager of all the records involving the water source.
Example – For Illustrative Purposes Only

WATER TEST FORM

<table>
<thead>
<tr>
<th>Date</th>
<th>Sample Point</th>
<th>Testing Lab</th>
<th>Analysis Results</th>
<th>Operator Initials</th>
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Reviewed by________________________________

Date ______________________________________

Date ______________________________________

Date ______________________________________
Example – For Illustrative Purposes Only

**HACCP PLAN**

**COMPANY NAME**
Small Fortune Aquaculture

**UNIT OPERATION**
Grow out

**CONTROL POINT**
Water (source)

**CRITICAL:** Yes or No
YES

**HAZARD OR DEFECT:**
Diseases and pathogens
Heavy metals
Pesticides
Bacteria
Industrial organics (PCBs)
PANO (Poly-Aromatic Nucleated Organics = petroleum decomposition products)

**CRITICAL LIMIT:**
Standards – establish limits based on end use of product (fish)

**PREVENTIVE MEASURES:**
Periodically analyze water
   Independent laboratory
   Appropriate tests
HACCP PLAN (Cont.)

MONITORING PROCEDURES

Twice annually – analyze water (source) for hazards
Twice annually – analyze fish for hazards

CORRECTIVE ACTIONS:

If water doesn’t meet specifications
   1. quarantine fish
   2. analyze fish for safety
   3. investigate water source for problem

RECORDS:

Records for every water sample
   1. date of sample
   2. sample point (location)
   3. where sample was analyzed
   4. results of analysis
   5. person taking sample

VERIFICATION PROCEDURE/PERSON(S) RESPONSIBLE:

Facility manager reviews records after each sample period
   – confirms records and sample accuracy
Records are reviewed by manager annually
Feed was also determined to be a critical control point. Possible hazards or defects include: mycotoxin, heavy metals, pesticides, herbicides, lipid rancidity, industrial organics (PCBs), and PANO (Poly-Aromatic Nucleated Organics = petroleum decomposition products). The critical limits will be established based on end use of the produce (fish) and on the sensitivity of the specific fish being grown. Preventive measures include periodically analyzing the feed using an independent laboratory to make sure the feed meets specifications and does not contain hazards or defects. The monitoring procedures include: on every new feed shipment – take a sample and store appropriately, twice annually have the feed analyzed by an independent lab, twice annually have the fish analyzed for hazards. Corrective actions include: 1. Impound the feed remaining, 2. Quarantine all fish fed this feed, 3. Analyze fish for safety, 4. Meet with feed manufacturer to resolve the problem. Records for feed should include (for every delivery): feed manufacturer, date of delivery, quantity and type of feed, date of manufacture and lot number, person taking the sample, sample storage identification, and results of analysis. The manager reviews records after each deliver to confirm accuracy and quarterly review all records on feed for verification.

Example – For Illustrative Purposes Only

**FEED FORM**

<table>
<thead>
<tr>
<th>Date</th>
<th>Sample ID</th>
<th>Lab ID</th>
<th>Mill</th>
<th>Lot No.</th>
<th>Quantity</th>
<th>Type</th>
<th>Protein</th>
<th>Analysis Results</th>
<th>Operator Initials</th>
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Reviewed by ___________________________

Date ________________________________
HACCP PLAN

Company Name: Small Fortune Aquaculture
Unit Operation: Grow out

CONTROL POINT
Feed: YES

HAZARD OR DEFECT:
Mycotoxin
Heavy metals
Pesticides
Bacteria
Herbicides
Industrial organics (PCBs)
PANO (Poly-Aromatic Nucleated Organics = petroleum decomposition products)

CRITICAL LIMIT:
Standards – establish limits based on end use of product (fish)

Example – PCBs – preferable 0.0 – limit of 1 ppb in edible fish

PREVENTIVE MEASURES:
Periodically analyze feed
    independent laboratory
    meets specifications
HACCP PLAN (Cont.)

MONITORING PROCEDURES

Every new feed shipment – sample and store
Twice annually – analyze feed
Twice annually – analyze fish for hazards

CORRECTIVE ACTIONS:

If feed doesn’t meet specifications or has hazard
   1. Impound feed
   2. Quarantine all fish fed this feed
   3. Analyze fish for safety
   4. Meet with feed manufacturer – resolve problem

RECORDS:

Records for every feed delivery
   1. feed manufacturer
   2. date of delivery
   3. quantity and type of feed
   4. date of manufacture and lot number
   5. person taking the sample
   6. sample storage identification
   7. results of analysis

VERIFICATION PROCEDURE/PERSON(S) RESPONSIBLE:

Facility manager reviews records after each delivery
   – confirms records and sample accuracy
Records are reviewed by facility manager quarterly
Within the grow out unit, other plants and animals will be growing as well. Many of these organisms are not harmful and some are indispensable to the operation of the system (nitrifying bacteria). Collectively these organisms are identified as flora and fauna in the flow chart. This control point is classified as critical. The hazards that have been identified are: algae and actinomycete, Listeria, Aeromonas, Salmonella, other bacteria, and certain parasites. These organisms are identified as a hazard since they can be contracted by humans and/or they will cause disease if eaten by human (raw or improperly cooked). The critical limits will be established based on the end use of the product (fish). Ornamental fish not intended for human consumption will have a different set of critical limits than will fish intended for the raw fish bars. Preventative measures include testing the fish and water periodically utilizing an independent laboratory performing appropriate tests. During the growth cycle (preferably just before harvest), the fish and water is analyzed for the specific hazards. Corrective actions that must be taken if either the fish or water does not meet specifications include: 1. Quarantine fish, 2. Analyze fish for safety, 3. Investigate source of the problem. Records for every sample taken should include: date of sample, where the sample was taken, the laboratory conducting the analysis, and the results of the analysis. The facility manager reviews the records after each sample period to confirm the records and sample accuracy. The entire set of records is review by the manager annually.

Example – For Illustrative Purposes Only

**FLORA AND FAUNA TEST FORM**

<table>
<thead>
<tr>
<th>Date</th>
<th>Sample Point</th>
<th>Testing Lab</th>
<th>Analysis Results</th>
<th>Operator Initials</th>
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Reviewed by_____________________________________

Date ___________________________________________
Example – For Illustrative Purposes Only

HACCP PLAN

COMPANY NAME
Small Fortune Aquaculture

UNIT OPERATION
Grow out

CONTROL POINT
Flora and Fauna

CRITICAL: Yes or No
YES

HAZARD OR DEFECT:
Algae an Actinomycete
Listeria
Aeromonas
Salmonella
Bacteria
Parasites

CRITICAL LIMIT:
Standards – establish limits based on end use of product (fish)

PREVENTIVE MEASURES:
Test fish and water periodically
Independent laboratory
Appropriate tests
HACCP PLAN (Cont.)

MONITORING PROCEDURES

During growth cycle
   analyze fish for hazards
   analyze water for hazards

CORRECTIVE ACTIONS:

If do not meet standards
   1. Quarantine fish
   2. Analyze fish for safety
   3. Investigate source of problem

RECORDS:

Records for every sample
   1. Date of sample
   2. Sample point
   3. Where sample was analyzed
   4. Results of analysis
   5. Person taking the sample

VERIFICATION PROCEDURE/PERSON(S) RESPONSIBLE:

Facility manager reviews records after each sample
   – confirms records and sample accuracy
Records are reviewed by manager annually
The sole output identified for Small Fortune Aquaculture is the harvest. This control point was identified as critical. Potential hazards include: off-flavors (food fish only), damage to fish, toxic substances, and pathogens. The critical limit standards will be established based on the end use of the product (fish). Preventive measures involve testing the fish prior to harvest for the potential hazards or defects identified as critical limits. Monitoring includes: during the growth cycle analyzing the fish for hazards and flavor testing (food fish only). Corrective actions would include quarantining the fish, analyzing for safety and investigating the source of the problem. Records include date of sample, unit sampled, weight of fish harvested, the testing laboratory used, the results of analysis, and the person taking the sample. The facility manager reviews the records after each sample to confirm the records and sample accuracy. All records of harvest are reviewed annually by the manager.

Example – For Illustrative Purposes Only

**HARVEST FORM**

<table>
<thead>
<tr>
<th>Date</th>
<th>Unit Harvested</th>
<th>Weight Harvested</th>
<th>Testing Laboratory</th>
<th>Results</th>
<th>Operator Initials</th>
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Reviewed by______________________________

Date _________________________________
Example – For Illustrative Purposes Only

**HACCP PLAN**

**COMPANY NAME**
Small Fortune Aquaculture

**UNIT OPERATION**
Grow out

**CONTROL POINT**
Harvest

**CRITICAL: Yes or No**
YES

**HAZARD OR DEFECT:**
Off-flavors (food fish only)
Damage to fish
Toxic substances
Pathogens

**CRITICAL LIMIT:**
Standards – establish limits based on end use of product (fish)

**PREVENTIVE MEASURES:**
Test fish prior to harvest
  Independent laboratory
  Appropriate tests
HACCP PLAN (Cont.)

MONITORING PROCEDURES

During growth cycle – analyze fish for hazards
Flavor test (food fish only)
Appropriate tests

CORRECTIVE ACTIONS:

If do not meet specifications
1. Quarantine fish
2. Analyze fish for safety
3. Investigate source of problem

RECORDS:

Records for every sample
1. Date of sample
2. Unit harvested
3. Weight of fish harvested
4. Testing laboratory used
5. Results of tests
6. Person taking samples

VERIFICATION PROCEDURE/PERSON(S) RESPONSIBLE:

Facility manager reviews records after each sample
– confirms records and sample accuracy
Records are reviewed by manager annually

Concluding remarks:
A HACCP plan will help insure that a safe and wholesome product is produced and sold to consumers. It can also be a valuable management tool for the owner/operator of a recirculating aquaculture facility. Although aquaculture producers are not required by law to have a HACCP plan, good facility and business management would dictate that a HACCP plan be used for maximum operating efficiency.
HACCP Program for Disease and Therapeutics for Intensively Cultured Foodfish

Stephen A. Smith  
Assistant Professor of Aquatic Animal Medicine/Fish Health  
Department of Biomedical Sciences and Pathobiology  
Virginia-Maryland Regional College of Veterinary Medicine  
Virginia Polytechnic Institute and State University

Introduction

The incorporation of a Hazard Analysis Critical Control Point (HACCP) system into the fish health program of an aquaculture facility is one way to help guarantee a safe and wholesome product for both the producer and consumer of cultured finfish. This system, like that used in seafood safety regulations, is based on the analysis of potential health hazards for the fish and human consumer, determination of times or locations where these hazards may occur during the life of the fish, and institute of measures for prevention or elimination of those hazards. Though a HACCP program for fish health is not technically required by federal regulations, the implementation of such a system would also help insure a consistent marketable product.

HACCP program for fish health

The first step of a fish health HACCP system is the identification of hazards and risks associated with the production, processing and consumption of the fish. These would include potential problems that may affect the health of either the fish being produced or the consumer who eats the product. Clinical and subclinical bacterial, viral and parasitic diseases, contaminants such as heavy metals, pesticides, herbicides and chemotherapeutics, and water treatment chemicals are all examples of hazards or risks that may affect the health of the fish or the health of the consumer who eats the contaminated product.

The next step is to determine critical control points where the hazards or risks to the fish and future product can be prevented or controlled. In addition, critical limits for each hazard or risk must be established for each identified critical control point. The majority of these critical limits will be established based on the end use of the product. For instance, most heavy metals, pesticides and herbicides have maximum limits for products intended for human consumption. This part of the HACCP program should also include developing procedures for monitoring or detecting the potential hazards or risks at the identified critical control points. Therefore, periodic collection and analysis of fish from the aquaculture facility for heavy metals, pesticides and herbicides would help insure that the end product is not contaminated with these substances. Regular monitoring will establish a baseline for such substances and will also help to identify major points or sources of contamination for the aquaculture facility. Another example of a hazard or risk to the final product is where there is contamination of the fish product with chemotherapeutics or water chemicals used by the aquaculture facility. This can occur with either FDA approved or unapproved chemical or chemotherapeutics. Again, regular monitoring
of the final product will help determine if there is any potential product contamination associated with their use.

Next, corrective measures must be established and monitored to correct any deviation from the previously established critical control point limits. Thus, depending on the type and amount of contamination, the fish or end product may have to be held until levels fall below the established critical limits, or the fish may have to be destroyed or the final product discarded. For example, fish contaminated with unacceptable levels of a FDA approved chemotherapeutic may only have to be held for an additional amount of time until the levels of that compound fall within acceptable levels. However, fish contaminated with an unapproved chemical or chemotherapeutic would most likely have to be destroyed and excluded from the human consumption market.

Finally, the hallmark of a successful HACCP program is the maintenance of an accurate record keeping system. All identified critical control points with their associated hazards or risks should be clearly outlined, as well as all critical limits, monitoring procedures, time schedules and possible corrective measures. This would involve keeping comprehensive records on all individual lots of fish including the source of the fish, data received, results of initial and regular diagnostic examinations, and a complete history of any problems or diseases encountered. Extensive records should also be kept for all water chemical and therapeutic treatments utilized including the data treated, the condition treated, the chemical or drug used, the source of the compound, the amount or dosage used, and the length of the treatment. Thus, only through the incorporation and full documentation of a HACCP program directed towards the health of the fish and ultimate end product can a producer insure a consistent, safe and wholesome product for both the producer and consumer of cultured finfish.

References
