Feed technology and fish nutrition

NOVA course
Lecture 14
Vegard Denstadli
Lecture outline

- Wide topic: will mainly use examples from salmonids and marine fish (carnivores)
- Special demands for fish feed
- The extruder line
- Nutrients and antinutrients – fate during processing
- Moist fish feed
- Pellet types
Special demands for fish

- High protein, high fat (>20% fat)
- Low tolerance for starch (low amylase activity) in salmonids (15 % is close to maximum)
- BUT: starch gelatinisation of granules is needed for good physical quality (covered by Birger)
- Low tolerance for CHO in general (unless a herbivore with long GI tract). No or little fermentation capacity.
- The pellet must be water stable
- The pellet must be able to absorb oil
- Oil or fat must have low melting point since fish are poikilothermic (fish temperature is the same as the ambient water)
Amylase activities in the liver and intestine of selected teleosts at 37 °C *(Hidalgo et al. 1999)*

<table>
<thead>
<tr>
<th></th>
<th>Liver</th>
<th>Intestine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carp</td>
<td>108</td>
<td>73</td>
</tr>
<tr>
<td>Goldfish</td>
<td>24</td>
<td>76</td>
</tr>
<tr>
<td>Tench</td>
<td>13.1</td>
<td>19.4</td>
</tr>
<tr>
<td>Seabream</td>
<td>2.7</td>
<td>1.8</td>
</tr>
<tr>
<td>Trout</td>
<td>0.0</td>
<td>1.3</td>
</tr>
</tbody>
</table>
The processing line

- Silo
- Dosing
- Grinding
- Mixing
- Conditioning
- Extrusion
- Drying
- Vacuum coating
- Cooling
Pre-conditioner and extruder

Conditioning + extrusion: < 5 minutes
Already covered

- Grinding
- Mixing
- Pre-conditioning
- Extrusion
- Vacuum coating
Grinding

- Most ingredients in fish feed mixtures are ground finely (<1mm screen) prior to extrusion
- Increased surface area
  - Improved gelatinisation
  - Improved physical quality
- Evacuation rate from stomach slower when feeding diets with large particle size (Sveier et al. 1999)
- Suppressed fat and energy digestibility when feeding coarse diets (FM based) (Sveier et al. 1999)
- No "muscle stomach grinding" as seen with poultry

Example

- Salmon
- 15% SBM inclusion in FM based diet
- Coarse, moderately (200μm) and fine (50μm)
- Fine grinding: Improved growth, and increased feed intake

Storebakken & Roem, 2004
Conditioning

- Add moisture and heat (water and steam) and initiate starch gelatinisation and protein denaturation (approx. 90°C)
- Reduce mechanical power consumption and increase extruder capacity
- Effect of moisture on starch gelatinisation:

![Graph showing gelatinisation (DSC) vs. retention time for different water contents (20%, 25%, and 30%). 30 s may be sufficient for fine ground meals.]

www.umb.no
## Effects of conditioning

<table>
<thead>
<tr>
<th>Processing method</th>
<th>Weight gain (g/fish)</th>
<th>FCR&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unground, no steam conditioning</td>
<td>54.5±5.5</td>
<td>2.01±0.04</td>
</tr>
<tr>
<td>Unground, steam-conditioned</td>
<td>71.3±3.2</td>
<td>1.75±0.04</td>
</tr>
<tr>
<td>Ground, no steam conditioning</td>
<td>61.3±2.1</td>
<td>1.91±0.09</td>
</tr>
<tr>
<td>Ground, steam-conditioned</td>
<td>70.7±1.5</td>
<td>1.76±0.02</td>
</tr>
<tr>
<td>Ground, extruded</td>
<td>58.2±4.9</td>
<td>1.50±0.01</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Processing method</th>
<th>Steam-conditioned</th>
<th>No steam conditioning</th>
<th>Extruded</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Digestibility coefficient</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry matter (%)</td>
<td>67.00±1.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>64.76±0.97&lt;sup&gt;a&lt;/sup&gt;</td>
<td>71.40±0.32&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Gross energy (Mj/kg)</td>
<td>77.95±1.10&lt;sup&gt;a&lt;/sup&gt;</td>
<td>76.47±0.73&lt;sup&gt;a&lt;/sup&gt;</td>
<td>83.22±0.22&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>90.16±0.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>88.97±0.49&lt;sup&gt;a&lt;/sup&gt;</td>
<td>89.28±1.62&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>
How can feed processing...

- **Positively** affect nutrient value of fish feed
- **Negatively** affect nutrient value of fish feed

Temperature
Water
Time
Nutrients - classification

- **Proteins**
  - Large protein molecules
  - Peptides

- **Lipids**
  - TAG (neutral fat)
  - PL (cell wall)

- **Carbohydrates**
  - Starch
  - Complex CHO (fibre)

- **Minerals**
  - Macro minerals
  - Micro minerals

- **Other**
  - Vitamins
  - Pigment
  - Enzymes
Fate of nutrients and antinutrients

- Starch gelatinisation
- Protein denaturation and unfolding

- Disulfide bonds formed
- Maillard rx: AA + reducing sugar
- Enzyme inactivation
- Loss of unstable vitamins

- Drying

Temperature ranges:
- 80-100°C
- 120-140°C
Figure 6: Temperature and retention time in the pre-conditioner, extruder and through the die (Reprinted with permission from Munch, 2002). The temperature increase in the pre-conditioner is basically caused by steam and moisture addition (STE). The temperature increase in the extruder is generated from mechanical dissipation (SME). Venting removes steam (and thus temperature) causing a drop in the temperature before the last section. Expansion of the pellet as the feed exits the die, cause moisture flash off and thus, removes heat.
Temperature profiles in the extruder barrel
However:

- Extrusion cooking normally does not induce severe damages to the feed as long as the mash is properly conditioned
  - Sørensen et al. (2002) tested extrusion at 100, 120 and 150 degrees and found no effects on nutrient digestibilities in salmon
  - Similar results obtained by Barrows et al. (2007)

- No major formation of disulphide bonds during conventional extrusion cooking
  - Disulphide (SS) bonds were assessed in FM and SBM diets (23-32 nmol/mg crude protein)
  - No effects of SS content on digestibility (Aslaksen et al. 2006)
Antinutrients

- Plant derived components that may negatively effect the utilisation of nutrients (proteins, lipids, starch)
  - Heat stable
    - Phytates
    - NSP, oligosaccharides
  - Heat labile
    - Protease inhibitors
    - Lectins

Feed enzymes post extrusion or enzyme pre-treatment
Trypsin inhibitors (TiA, mg/kg)

Soy bean meal diet

White flakes diet

Before extrusion

After extrusion

Romarheim et al. 2006
Antinutrients

Table 2: Effect of temperature on trypsin inhibitory activity

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Extrusion temperature °C</th>
<th>Trypsin Inhibitory units mg/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>40</td>
<td>51*</td>
</tr>
<tr>
<td>2.</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>3.</td>
<td>60</td>
<td>45</td>
</tr>
<tr>
<td>4.</td>
<td>70</td>
<td>41</td>
</tr>
<tr>
<td>5.</td>
<td>80</td>
<td>34</td>
</tr>
<tr>
<td>6.</td>
<td>90</td>
<td>21</td>
</tr>
<tr>
<td>7.</td>
<td>100</td>
<td>14</td>
</tr>
<tr>
<td>8.</td>
<td>110</td>
<td>9</td>
</tr>
<tr>
<td>9.</td>
<td>120</td>
<td>5</td>
</tr>
<tr>
<td>10.</td>
<td>125</td>
<td>&lt;2</td>
</tr>
<tr>
<td>11.</td>
<td>130</td>
<td>&lt;2</td>
</tr>
<tr>
<td>12.</td>
<td>135</td>
<td>&lt;2</td>
</tr>
<tr>
<td>13.</td>
<td>140</td>
<td>&lt;2</td>
</tr>
</tbody>
</table>

*for raw Soya beans
Lectin inactivation

![Graph showing lectin inactivation at different temperatures](chart.png)
Vitamins

- Processing of feed ⇒ loss of vitamin E, due to lability to heat.
- Loss of naturally occurring vit E from ingredients.
- Must be added in stable form:

- Vitamin E: alphatocopherol acetate
- Vitamin C: StayC (polyphosphate)
Feed enzymes

- Only feed enzyme allowed for fish feed in EU is phytase
- Cannot be added directly in mash prior to conditioning and extrusion due to heat instability (although novel phytases are more stable, and can tolerate pelleting temperatures, extrusion temperatures are still too high)

- Applications
  - vacuum coater (must be emulsified in oil)
  - Pre-treatment
What about 120-140°C?

(Brinch-Pedersen et al., 2006)
Know your enzyme:

- Storage stability (before and after processing)
- Optimal
  - pH
  - Temperature
- Stability during feed processing
- Does it resist being digested by intestinal proteases when *in vivo*?
- When is it active in the GI tract?
- Two main types
  - Granulate/powder
  - Liquid
pH

• Most feed ingredients (feed mash)
  • pH 6-7
• GI tract
  • Pig:

Depending on the "broadness", the enzyme must be designed for acidic or neutral/alkaline conditions
pH in fish

- pH 4-5: Stomach
- pH 7-9: Other areas
Thermodependent activities of two xylanases and one glucanase

Collins et al. 2005
Thus: Addition of feed enzymes in diets for coldwater fish is suboptimal.

Alternative: Online enzyme pre-treatment of (vegetable) feed ingredients at optimal conditions for the enzyme
Phytase

IP$_6$

Lim et al., 2000
Online incubation with phytase
Phytase pretreatment

Denstadli et al., 2006
Pellet quality

- A prerequisite is acceptable physical quality for transport and feeding systems
- However: the pellets cannot be overprocessed to in order to obtain e.g. good durability. They need to be digested in the end
- Typical parameters that are measured:
  - Amount of dust
  - Durability
  - Hardness
  - Water stability
  - Loss of fat/fat leakage
Effects of high and low pellet water stability

Bæverfjord et al., 2006
Pellet appearance after 240 min with moderate shaking

- High WS
- Low WS
Moist feed

- Annually unutilised marine by-products: 20-25 million tonnes

- Challenges
  - Logistics
  - Storage stability
  - Feed waste (pellet quality)

- Positive effects
  - Good growth and quality
  - Resource utilisation

(RUBIN, 2000)
GellyFeed® – alkaline preservation

Alkaline fish silage – step one

GellyFeed – step two

Byproducts → Mincer → Base

Alkaline fish silage pH >11

3% Dry ingredients

Mixer → Pelleting unit → Pellet (Water-stable)

Sørensen and Denstadli, 2008
The GellyFeed process

- Addition of base (KOH) inactivates enzymes, and inhibits proteins autolysis
- The alkaline silage (60-70% moisture) is mixed with dry ingredients (including seaweed binder, alginate)
- The final structure of the feed is set by immersing the pellet in a formic acid (CH₃COOH) bath for 24 h, and the final pH was adjusted to pH 4.5–5.
- Binding in the pellet takes place due to alginates ability to form gel in acidic environment, in the presence of Ca²⁺ ions
- Final dry matter: 35%
GellyFeed® – alkaline preservation
Diet formulation and digestibility of Gellyfeed in salmon (3kg)

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Digestibility, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat</td>
<td>96</td>
</tr>
<tr>
<td>Protein</td>
<td>81</td>
</tr>
<tr>
<td>Energy</td>
<td>87</td>
</tr>
</tbody>
</table>

| Preserved herring by-products (g kg\(^{-1}\))    | 965              |
| Alginate (g kg\(^{-1}\))\(^a\)                   | 33               |
| Premix of vitamins and minerals (g kg\(^{-1}\))\(^b\) | 2                |
| Carophyll pink (mg kg\(^{-1}\))\(^c\)            | 150              |
| Yttrium oxide (mg kg\(^{-1}\))                   | 50               |
Growth in salmon fed Gellyfeed and dry extruded feed

<table>
<thead>
<tr>
<th></th>
<th>Moist feed</th>
<th>Control feed</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start weight (g)</td>
<td>1648 ± 36.0</td>
<td>1720 ± 2.8</td>
<td>NS</td>
</tr>
<tr>
<td>Final weight (g)</td>
<td>3085 ± 31.2</td>
<td>2912 ± 27.2</td>
<td>0.014</td>
</tr>
<tr>
<td>Weight gain (g)</td>
<td>1438 ± 34.0</td>
<td>1193 ± 21.0</td>
<td>0.004</td>
</tr>
<tr>
<td>Specific growth rate (%)</td>
<td>0.53 ± 0.015</td>
<td>0.44 ± 0.012</td>
<td>0.012</td>
</tr>
<tr>
<td>Thermal growth coefficienta</td>
<td>3.1 ± 0.08</td>
<td>2.6 ± 0.05</td>
<td>0.005</td>
</tr>
</tbody>
</table>
Sinking velocity

- Depends on the feeding behaviour of the species
  - Floating
  - Slow sinking
  - Fast sinking

- Sinking velocity is affected by:
  - Pellet expansion after leaving the extruder die
  - Oil content
  - Fresh water, brackish water or saltwater (3.4% NaCl)

- A simple way to regulate this by the operator is to increase or reduce steam venting in the 3rd/4th section of the extruder barrel

- Measure bulk density directly after the die (this is app. The same as after the dryer since the pellets shrink as they loose moisture in the dryer
Bulk densities and sinking behaviour

<table>
<thead>
<tr>
<th></th>
<th>Bulk density (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fast sinking</td>
<td>640-680</td>
</tr>
<tr>
<td>Slow sinking</td>
<td>600-630</td>
</tr>
<tr>
<td>Neutral buoyancy in salt water</td>
<td>570-590</td>
</tr>
<tr>
<td>Neutral buoyancy in freshwater</td>
<td>550-570</td>
</tr>
<tr>
<td>Floating</td>
<td>520-540</td>
</tr>
</tbody>
</table>

Pellet density before coating (g/l) = \( \frac{\text{Final pellet density (g/l)} \times [100 - \text{final fat %}]}{[100 - \text{initial fat %}]} \)
Pellet size
Species differences

(Einen and Mørkøre, 1996)
Salmonids (salmon, trout)

- First feeding:
  - Directly on formulated feed

- Size
  - Start feeding: 0.5 mm
  - Grow out: up to 12 mm

- Sinking velocity
  - Slow sinking pellets
Marine fish (cod)

- First feeding: rotatoria (artemia)
  - Size
    - Larval feeding: 75 micron
    - Start feeding: 0.5 mm
    - Grow out: up to 13 mm
  - Appearance in water
    - Slow sinking pellets

- Formulated microgranules
  - Highly available nutrients
    - n-3 fatty acids
    - Peptides, amino acids
    - Vitamins, minerals
    - Nucleotides
  - Appearance after 5-15 days