Controlling Microbiological Pathogens in Brine Systems

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WAFF Fall Food Safety Workshop
November 2, 2017
Agenda

• Microbiological quality of brines
• Antimicrobial additives for brines
• Other control methods
• Question & Answer
Microbiological Quality of Brines

• **Cheese Brines**
  
  • During brining, NaCl molecules move as ions from the brine to the curd due to the difference in osmotic pressure.
  
  • Over time, the brine composition changes with salt being exchanged for proteins, fats, sugars, lactate and minerals.
  
  • Use of brines over long periods of time can lead to significant increases in microflora which can lead to cheese defects like mold growth, pigment spots and cheese softening due to excessive proteolysis.
  
  • Pathogens can also survive in brines for several weeks.
Microbiological Quality of Brines

• **Cheese Brines**
  
  • Complete replacement of old brines is expensive
    • Cost of materials - water and salt
    • Disposal of old brine
  
  • Fresh brines can cause cheese to be slimy and sticky, referred to as “soft-rind” or “rind-rot”.
  
  • Yeast present in cheese brines can raise the pH and make it more hospitable for pathogens.
  
  • Brine regeneration is therefore preferred and restores the chemical and microbiological quality of the brines.
Microbiological Quality of Brines

• Meat Chilling Brines
  • Source of *Salmonella* or *Campylobacter* cross-contamination in raw poultry.
    • Rolling 52 week window for whole birds and parts
      • Must stay below a certain percentage i.e., no more than 7.1% of turkey carcass can be positive for *Salmonella* or will be subject to further testing or regulatory action.
  • Source of spoilage or pathogens for RTE meat products.
    • Zero tolerance for *Listeria monocytogenes*
  • Again, it’s expensive to dispose of and replace spent brines…and so far they are not using spent meat brines on the road!
Antimicrobial Additives for Brines
Chlorine (cheese brine and meat chilling)

- **One of the most widely used sanitizers**
  - Up until 2010, chlorine was the most common processing aid in poultry chillers.
  - Limited to 50 ppm per USDA and 10-100 ppm for cheese brine but up to 400 ppm has shown to be necessary to be effective in poultry chill waters.

- **Inexpensive**

- **Exhibits broad spectrum antimicrobial activity**

- **Active chlorine acts on cell membranes, inhibits cellular enzymes, attacks DNA and oxidizes cellular proteins**
Chlorine (cheese brine and meat chilling)

Depending on the pH of the solution, the active chlorine from hypochlorite solutions exists as three primary species.

Never Mix Acids With Chlorine!!!
Chlorine (cheese brine and meat chilling)

In alkaline solutions, the predominate species is the hypochlorite ion.

- Hypochlorite is a strong oxidizer
- But it has difficulty penetrating the lipid bilayer
- Causes damage only to outer structures of the cell
Chlorine (cheese brine and meat chilling)

In acid conditions the hypochlorous acid increases

- Hypochlorous acid can pass through the cell membrane
- Attacks vital internal structures such as DNA as well as structures on the outside of the cell
- This is much more lethal to the bacteria

For this reason, hypochlorous acid is a more effective sanitizer than the hypochlorite ion.
Chlorine (cheese brine and meat chilling)

• It’s inexpensive but…
  • Corrosive to softer metals like aluminum
  • Efficacy decreases at higher pH levels
  • Effectiveness is reduced at lower temperatures typical of cooling brines for meat and cheese.
  • Efficacy is decreased in the presence of organic matter.
  • Chlorine levels autonomously decrease as a function of initial concentration, time, and temperature.
Peroxyacetic Acid (PAA) - Meat only

- Consists of equilibrium solutions of PAA, hydrogen peroxide, and acetic acid
- Broad spectrum antimicrobial activity, including yeast, molds, algae, and spores.
- Works well at lower temperature
- Effective up to pH 8.5
Peroxyacetic Acid (PAA) - Meat only

- PAA’s are one of the most effective agents against microorganisms and biofilms because of their powerful oxidizing properties.

![Diagram of Hydroxyl radical](image)
Peroxyacetic Acid (PAA) - Meat only

• Mode of Action:
Peroxyacetic Acid (PAA) - Meat only

• Oxidizes proteins, lipids, and nucleic
• Leads to disruption of the cell membrane which causes cell lysis and microbial death.
• Breaks down to form the non-harmful compounds
  • Acetic acid
  • Water
  • Oxygen
• Approved for use in poultry chilling waters and RTE meat products
Peroxyacetic Acid (PAA) - Meat only

- PAA is used by the majority of poultry processors in the US.

<table>
<thead>
<tr>
<th>Chill Water Treatment</th>
<th>Carcass Sampling Point</th>
<th>Salmonella % Positive</th>
<th>% Reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>85 ppm PAA</td>
<td>Pre-Chill</td>
<td>30.5</td>
<td>91.8</td>
</tr>
<tr>
<td></td>
<td>Post-Chill</td>
<td>2.5</td>
<td></td>
</tr>
<tr>
<td>30 ppm Chlorine</td>
<td>Pre-Chill</td>
<td>25.5</td>
<td>56.8</td>
</tr>
<tr>
<td></td>
<td>Post Chill</td>
<td>11.0</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chill Water Treatment</th>
<th>Carcass Sampling Point</th>
<th>Campylobacter % Positive</th>
<th>% Reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>85 ppm PAA</td>
<td>Pre-Chill</td>
<td>83.0</td>
<td>43.4</td>
</tr>
<tr>
<td></td>
<td>Post-Chill</td>
<td>47.0</td>
<td></td>
</tr>
<tr>
<td>30 ppm Chlorine</td>
<td>Pre-Chill</td>
<td>78.0</td>
<td>12.8</td>
</tr>
<tr>
<td></td>
<td>Post Chill</td>
<td>68.0</td>
<td></td>
</tr>
</tbody>
</table>

Bauermeister, Laura J. Evaluation of Poultry Meat Safety and Quality using Peracetic Acid in Poultry Chillers. 2015
Ozonation (cheese brine and meat chilling)

- **Ozone is generated on site by exposing oxygen to high voltage electricity.**
  - This is then bubbled into the brine/water and the ozone breaks down into hydrogen peroxy (HO$_2$) and hydroxyl (OH) each of which strong oxidizers.
- **Environmentally friendly**
  - Decomposes naturally and components are released as oxygen, leaving behind no chemical residues.
  - Considered GRAS by FDA
Ozonation (cheese brine and meat chilling)

- **Ozone is a strong oxidizing agent, stronger than chlorine and PAA.**
  - Oxidizes and destroys the cell wall and cell membrane leading to cell lysis.
  - Also attacks enzymes and DNA
  - 2 ppm is the max allowable

- **However...**
  - The equipment is expensive
  - Ozone is extremely irritating so off gases must be monitored and eliminated to prevent worker exposure.
Ozonation (cheese brine and meat chilling)

Fig. 2. Effect of ozone treatment at 0.40 mg L⁻¹ for different times (0–240 min) on log counts (▲, total viable count; ■, microstaphylococci; □, yeasts) in the six brines used (B1–B6). Error bars represent standard deviation; regular lines represent data fitted by Weibull model.

Hydrogen Peroxide (cheese brine)

- **Compound with two hydrogen atoms and two oxygen atoms** ($\text{H}_2\text{O}_2$)
  - Similar to water but the extra oxygen atom is readily shed and can react with its environment and is destructive to microorganisms.
  - The destructive oxygen atoms have an unpaired electron which is unstable and extremely reactive also called a **free radical**.
  - Destroys the microorganism’s cell wall and allows its insides to leak out.
  - Leaves no residual chemicals behind
Hydrogen Peroxide (cheese brine)

- Effectiveness:

Chlorine dioxide (cheese brine and meat chilling)

- Generated by mixing sodium chlorite with acid
- Strong oxidizing agent
  - Attacks cell wall/membrane, enzymes, lipids, DNA
  - Better oxidizer than chlorine but less than PAA or ozone
- Must be generated on site as you can not transport the gas in the US.
- It is toxic so human exposure must be monitored where used.
- Studies in meat chilling brines and cheese brines show poor efficacy in eliminating *L. monocytogenes* from brines due to divalent Ca ions and organic matter.
Bioionix (meat and cheese)

- Bioionix creates oxidizing disinfectants from water and salt, using a low voltage electrical field and catalytic plates.
  - Hydrogen peroxide, chlorine, and oxygen then go to work as the **hydroxyl radical**, a powerful oxidant
  - Hypochlorous acid also forms as a residual sanitizer so it is continuously cleaning itself and preventing biofilms from forming
Bioionix (applications for meat and cheese)
Bioionix (applications for meat and cheese)

• Pros
  • Environmentally friendly
    • Reduces chloride discharge in wastewater.
  • No organoleptic effect on products
  • Process can be fully automated and has real time monitoring
  • Works well even in turbid solutions
    • Will flocculate fats and proteins so they can be skimmed off

• Cons:
  • Expensive - Each installation is unique to the customer
<table>
<thead>
<tr>
<th>Sanitizer</th>
<th>Activity</th>
<th>Oxidation Capacity</th>
<th>Concentration</th>
<th>Effectiveness</th>
</tr>
</thead>
<tbody>
<tr>
<td>PAA</td>
<td>Oxidant</td>
<td>1.81</td>
<td>Up 80 ppm</td>
<td>pH 1-8, less sensitive to organic matter</td>
</tr>
<tr>
<td>Hypochlorites</td>
<td>Oxidant</td>
<td>1.36</td>
<td>1-3 ppm for rinsing</td>
<td>pH 6-7, sensitive to organic matter</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>50 ppm for sanitizing</td>
<td></td>
</tr>
<tr>
<td>Chlorine Dioxide</td>
<td>Oxidant</td>
<td>1.57</td>
<td>Up to 5 ppm</td>
<td>pH 6-10, less sensitive to organic matter</td>
</tr>
<tr>
<td>Hydrogen Peroxide</td>
<td>Oxidant</td>
<td>1.8</td>
<td>0.5%</td>
<td>Sensitive to organic matter</td>
</tr>
<tr>
<td>Ozone</td>
<td>Oxidant</td>
<td>2.07</td>
<td>2 ppm</td>
<td>pH 6-8, sensitive to organic matter, breaks down to $O_2$ rapidly</td>
</tr>
<tr>
<td>UV Light</td>
<td>Disrupts genetic material</td>
<td>-</td>
<td>40,000 µw s/cm²</td>
<td>pH independent, sensitive to organic matter</td>
</tr>
</tbody>
</table>

Other Interventions for Brines
UV Light (cheese and meat brine applications)

- Shortwave ultraviolet light (~254 nm) is a simple way to kill microorganisms.
- UV light attacks nucleic acids and disrupts their DNA, leaving them unable to perform vital cellular functions.
- Chemical free way to clean brines
UV Light

• Effectiveness is time dependent and measured as “microwatt seconds per square centimeter”
  • UV light does not penetrate very far into the solution and lamps can become fouled with organic matter, requiring a wiper system to clean the lamps.
  • Low temperatures can affect efficiency
  • One study showed a LM 5 log cfu/ml reduction in 45 minutes in spent brine but fresh brine only took 15 minutes (Parikh, P. et al. 2012. Control of LM in recycled chill brine using ultraviolet light and antimicrobial agents)
Microfiltration (meat and cheese applications)

- Membranes are typically made of natural or synthetic polymers that are coated with an inorganic substance like ceramic.
  - Pore sizes range from 0.1 to 20µm
- Removes microorganisms and physical contaminate from brine
  - No changes to the chemical composition of the brine but may need pre-filtration to remove fines to prevent membrane fouling.
  - Can continuously filter 5-20% of the total brine volume
Microfiltration

• Is not pH dependent
• Not temperature dependent
• Must clean the system to allow for optimal membrane functionality (CIP)
• High cost may make this difficult for small plants to implement.
Conclusions

• Many different options to “clean” your brines
• Factors to consider
  • Cost
  • Environmentally friendly
  • Efficacy
  • Peace of Mind…
Any Questions?

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