Stabilized Chlorine Dioxide in Fuel Ethanol Fermentation: Efficacy, Mechanisms and Residuals

XCELIS™

Derrick Okull, PhD
Technical Service Manager
Dupont Nutrition and Biosciences
Stabilized Chlorine Dioxide (ClO₂) – What is it?

- **NOT** chlorine/bleach
- Buffered sodium chlorite solution, antibiotic-free aqueous solution
- Historically used in food processing/water treatment industries for contamination control/sanitation
- **14yr history in fuel ethanol** – GRAS status, with expert concurrence, for use as a processing aid in potable alcohol and fuel ethanol production with resulting distillers’ grains approved for use in animal feed (cattle, swine, poultry).
- Controls lactic and acetic acid producing bacterial contamination
- Suitable for ethanol production from sugar and/or grain substrates
- Generated, concentrated solutions traditionally used in paper bleaching

<table>
<thead>
<tr>
<th>Property</th>
<th>Value/Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crystallization Point</td>
<td>-12°C 10°F</td>
</tr>
<tr>
<td>Freezing Point</td>
<td>-18°C 0°F</td>
</tr>
<tr>
<td>Boiling Point</td>
<td>106°C 223°F</td>
</tr>
<tr>
<td>pH</td>
<td>9.5 to 9.8</td>
</tr>
<tr>
<td>Density (g/mL) @ 20°C</td>
<td>1.18 to 1.21</td>
</tr>
<tr>
<td>Color</td>
<td>Clear light yellow solution</td>
</tr>
<tr>
<td>Odor</td>
<td>Faint chlorine-like smell</td>
</tr>
</tbody>
</table>
Stabilized Chlorine Dioxide – how it works

- A solution of 15 – 25 % Sodium Chlorite
- Chlorine dioxide is formed under acidic pH conditions
- When applied correctly, inactivates bacteria without harming yeast
- Activity is pH dependent – Need pH < 5.4 to be effective

Chlorite + Acid $\rightleftharpoons$ Chlorous acid + Chlorine dioxide

$\text{ClO}_2^- + \text{H}^+ \rightarrow \text{HClO}_2 + \text{ClO}_2$

Chlorite + Low pH mash/other substrate

Low pH/Acidic conditions drive conversion of Chlorite into more active form to kill bacteria on contact
Bacterial Contamination in the Ethanol Process:

A Primer
ETHANOL PRODUCTION - DRY GRIND PROCESS OVERVIEW

- Grinding/Slurry/Cook
- Liquefaction
- Fermentation
- Distillation
- DDGS
- Back Set
- Syrup
- Ethanol
- CO₂
- Corn Oil
WHERE DOES CONTAMINATION COME FROM?

- **Additives**

**WHERE DOES CONTAMINATION COME FROM?**

- CO2
- Beer well
- Distillation
- Liquefaction*
- Heat exchangers
- Cook
- Fermentation ~92°F, 60hrs
- Yeast propagation

- Dry ground corn
- Mash headers/piping/deadlegs

- Conducive temperature – 88 – 92°F
- pH – 4.9 – 5.6
- Food source – sugar, proteins, and trace/micronutrients
- Moisture – mash
- Protection - biofilms

**Microbiological Sampling of Various Fuel Ethanol Operations**

<table>
<thead>
<tr>
<th>Location</th>
<th>Typical Level of Lactic Acid Bacteria (CFU/ug)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Slurry Tank</td>
<td>100</td>
</tr>
<tr>
<td>Liquefaction Tank</td>
<td>100</td>
</tr>
<tr>
<td>Pre-heat exchanger</td>
<td>100</td>
</tr>
<tr>
<td>Post-heat exchanger</td>
<td>10000000</td>
</tr>
<tr>
<td>Early Fermentation (~15 h)</td>
<td>10000</td>
</tr>
<tr>
<td>Mid Fermentation (15-30 h)</td>
<td>1000</td>
</tr>
<tr>
<td>Late Fermentation (~30h)</td>
<td>1000</td>
</tr>
<tr>
<td>Yeast Propagation</td>
<td>100</td>
</tr>
</tbody>
</table>

**Drop lactic acid in a contaminated process**
WHY DOES BACTERIA CAUSE LOSSES?

- Competition with inoculant yeast for nutrients
- Increased production of organic acids (lactic and acetic) by contaminant bacteria instead of ethanol
- Inhibition of yeast
- Recycle through backset can also compound inhibition
- Downtime, cleaning costs from fouling as a result of unfermented sugar going into distillation
Controlling Contamination

**Process Controls**
- CIP/ Sanitation
- Temperature
- High Solids
- High Yeast inoculum
- pH Control/ Acids

**Antimicrobials**
- Hop Acids
- Oxidatives (eg ClO₂, H₂O₂, iodine, HOCI)
- Quaternary Compounds
- Glutaraldehyde
- Bacterial Peptides

**Antibiotics**
- Virginiamycin (Lactrol™)
- Monensin
- Erythromycin
- Penicillin
- Combinations

Cellular Targets of some common disinfectants

Cellular Targets of antibiotics

http://www.connect2cleanrooms.com/news-article/Rotational-Cleaning

Copyright © 2019 Dupont   All Rights Reserved
Example Plant Data
Lactic acid levels in a process before and after addition of ClO₂
ClO₂ Plant Performance

ClO₂ was trialed in conjunction with antibiotics at a 100 MGPY Dry Grind Ethanol Plant experiencing an active infection resulting in elevated lactic acid levels, high drop sugars, and decreased ethanol yields.
Dosing with ClO$_2$ helped to reduce the contamination in the system and move the plant back towards baseline profiles than treatment with antibiotics alone.
Dosing with \( \text{ClO}_2 \) helped to decrease lactic acid concentration in the system and move yields and sugar data back to baseline.
**Product Trail**

- **Engineering controls in place to limit exposure potential to workers**
  - Sodium Chlorite (NaClO₂)
  - H₂O
  - Other Ingredients

**Blending Process at Toll Manufacturing facility**

- **Transport to customer by tote**
  - Stable Product
  - Packaging/Storage/Transportation

**Possible Dosing Points**

- **Customer storage**

**Customer Process**

- **Grain Feedstock** (Corn/Wheat)
  - Milling
  - Liquefaction
  - Yeast Propagation
  - Fermentation
  - **Distillation**
    - **Dehydration**
    - **CO₂**
      - Atm.

- **Solids Separation**
  - Evaporation
  - **DDGS**
  - **Distillers Corn Oil**
  - **Transfer to Animal Feed producer**
    - **Blended Animal Feed**

**Proven safe (GRAS status) and approved for use in production of potable alcohol and fuel ethanol production with co-products intended for use in animal feed.**

Customer must receive guidance on recommended material safe handling, PPE and storage requirements.
### Residuals in DDGS

<table>
<thead>
<tr>
<th></th>
<th>Vol</th>
<th>ClO₂ dose (ppm)</th>
<th>Vol of FXL (gal)</th>
<th>Wgt (kg)</th>
<th>NaClO₂ (kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Preferm vol</td>
<td>180000</td>
<td>1000</td>
<td>180</td>
<td>720</td>
<td>159</td>
</tr>
<tr>
<td>Ferm 1</td>
<td>387000</td>
<td>1000</td>
<td>387</td>
<td>1548</td>
<td>341</td>
</tr>
<tr>
<td>Total working vol</td>
<td>1211000</td>
<td>468.2</td>
<td>567</td>
<td>2268</td>
<td>499</td>
</tr>
</tbody>
</table>

**Assumptions:**
- Continuous fermentation – vessels in series
- Dosing in the first two vessels in the train at up to 1000ppm (0.1% of working volume of dosed vessels)
- At drop, dilution has occurred across the system – total working volume includes all vessels in the train
- Stabilized ClO₂ contains 15-22% sodium chlorite (NaClO₂)
- At this dose, the system will have an estimated 499kg of chlorite
- End products of reaction in acidic environment are NaCl (salt) and NaClO₃ (chlorate) at about a 2:1 ratio, with the chlorate getting further reduced to NaCl
Typical Sodium in DDGS ~ (2000 ppm) 0.2%
Potential from high dose ClO₂ – 155 ppm (0.0155%)
Thank you!