Effect of supplemental vitamin C on meat quality of cattle fed varying concentrations of dietary sulfur

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Introduction

• Distillers grains are a nutritional and inexpensive alternative to corn
  ▫ Distillers grains can be high in sulfur (S) and fat
  ▫ Ranges include:
    • 4 - 12% fat (Stock et al., 2000)
    • S varies depending on plant
      • Can be over 1% S
Introduction

• High dietary S results in:
  ▫ Negative impacts on feedlot performance and meat quality
    • 0.6% S decreased ADG, HCW
    • Sulfur toxicity (PEM)
    • Oxidative stress
Introduction

• Acceptance of meat products by consumers primarily related to tenderness and flavor/marbling (Miller et al., 1995)

• Oxidative environment post mortem:
  • Oxidation of protease μ-calpain can limit the extent of muscle proteolysis = ↑ toughness
  • Oxidation of lipids = off flavors = ↓ shelf-life
Oxidation Slows Proteolysis and Tenderization

Intact Troponin-T

Troponin-T Degradation Product

Kg Shear Force

<table>
<thead>
<tr>
<th></th>
<th>C_{17}</th>
<th>E_{18}</th>
<th>C_{19}</th>
<th>E_{20}</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ox</td>
<td>Ox</td>
<td>Ox</td>
<td>Ox</td>
<td>Ox</td>
</tr>
<tr>
<td>Non</td>
<td>Non</td>
<td>Non</td>
<td>Non</td>
<td>Non</td>
</tr>
</tbody>
</table>

Oxidation Influences Autolysis of μ-Calpain

Effect of oxidation on progression of autolysis of μ-Calpain

<table>
<thead>
<tr>
<th></th>
<th>5 min</th>
<th>10 min</th>
<th>30 min</th>
<th>60 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>control</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ox</td>
<td></td>
<td>Ox</td>
<td>Ox</td>
<td>Ox</td>
</tr>
<tr>
<td>Reduced</td>
<td></td>
<td>Reduced</td>
<td>Reduced</td>
<td>Reduced</td>
</tr>
</tbody>
</table>

Ox = pre exposure to 200 µM H2O2;
Reduced = pre exposure to 0.1 % MCE

Lametsch, Huff-Lonergan and Lonergan. BBA Proteins and Proteomics, 1784:1215
Vitamin C Research

- Cattle synthesize ascorbate (vitamin C) in the liver
  - Roles in the body include:
    - Antioxidant function (redox reactions)
      - Regeneration of glutathione and vitamin E
    - Synthesis of collagen and carnitine
    - Enzyme cofactor
• In vivo
  • Japanese long fed cattle (Takahashi et al., 1999; Ohashi et al., 2000)
    ▫ Depletion of plasma vitamin C during finishing period
    ▫ Vitamin C added late in the finishing period increased marbling scores
    ▫ Jugular infusions of vitamin C improved color and lipid stability

• Cell culture work
  • Vitamin C enhanced adipocyte differentiation
    (Kawachi, 2006)
Study Objectives

• Determine the effects of supplemental vitamin C in diets containing varying concentrations of dietary S on:
  1. Performance of cattle
  2. Carcass characteristics of cattle
  3. Meat quality (protein degradation and oxidation)
120 steers, 5 pens/trt with 4 steers/pen
6 treatments

- Low Sulfur, 0.2% S
  - No Vitamin C
  - + Vitamin C

- Medium Sulfur, 0.4% S
  - No Vitamin C
  - + Vitamin C

- High Sulfur, 0.6% S
  - No Vitamin C
  - + Vitamin C

Rumen-protected vitamin C source included at 10g of vitamin C/head/day
Diet composition and analysis

<table>
<thead>
<tr>
<th>Item</th>
<th>Low S</th>
<th>Medium S</th>
<th>High S</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn</td>
<td>69.7</td>
<td>48.0</td>
<td>48.0</td>
</tr>
<tr>
<td>Corn dried distillers grains</td>
<td>18.0</td>
<td>40.0</td>
<td>38.9</td>
</tr>
<tr>
<td>Chopped hay</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
</tr>
<tr>
<td>Limestone</td>
<td>2.0</td>
<td>2.0</td>
<td>2.0</td>
</tr>
<tr>
<td>Salt</td>
<td>0.31</td>
<td>0.31</td>
<td>0.31</td>
</tr>
<tr>
<td>Vitamin A premix$^1$</td>
<td>0.10</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Trace mineral premix$^2$</td>
<td>0.035</td>
<td>0.035</td>
<td>0.035</td>
</tr>
<tr>
<td>Rumensin90$^3$</td>
<td>0.016</td>
<td>0.016</td>
<td>0.016</td>
</tr>
<tr>
<td>Sodium sulfate</td>
<td>--</td>
<td>--</td>
<td>1.11</td>
</tr>
<tr>
<td>Urea</td>
<td>0.8</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>S, %</td>
<td>0.18</td>
<td>0.32</td>
<td>0.56</td>
</tr>
<tr>
<td>CP, %</td>
<td>14.3</td>
<td>16.6</td>
<td>16.6</td>
</tr>
<tr>
<td>NEg, Mcal/kg DM</td>
<td>1.3</td>
<td>1.3</td>
<td>1.3</td>
</tr>
</tbody>
</table>

$^1$Vitamin A premix contains 4,400,000 IU/kg

$^2$Provided per kg of diet: 30 mg Zn as ZnSO$_4$; 20 mg Mn as MnSO$_4$; 0.5 mg I as Ca(IO$_3$)$_2$(H$_2$O); 0.1 mg Se as Na$_2$SeO$_3$; 10 mg Cu as CuSO$_4$; and 0.1 mg Co as CoCO$_3$

$^3$Provided at 27 g/ton diet
Study Timeline

Cattle received, step-ups

Day 0: weights, blood, liver, gas cap

Day 14: blood and gas cap

Day 28: weights, blood, and gas cap

Day 56: weights

Day 84: weights

Day 90: blood and gas cap

Day 112: weights

Day 143: blood, liver, gas cap

Day 148: weights

Sept: Study begins, limit fed, 7 d

Oct: Day -3 and -4: Ultrasound

Nov: Day 90 and 91: Ultrasound

Dec: Study ends, cattle harvested

Mar: 2011
Measurements taken

- **Live animal:**
  - BW, DMI, ADG, G:F, plasma vitamin C, plasma antioxidant capacity

- **Carcass:**
  - HCW, back-fat, KPH, REA, YG, QG, marbling score

- **Meat Quality**
  - Proximate analysis
  - Troponin-T degradation-Western Blotting
  - μ-Calpain autolysis-Western Blotting
  - Protein carbonylation-Oxi-Blots
  - Iron analysis-ICP-OES
  - TBARS
  - Fatty acid profile-GC-MS
Contrast Statements:
- A = Vitamin C vs. No Vitamin C
- B = Linear effect of Sulfur
- C = Vitamin C within High Sulfur
- D = Vitamin C within Sulfur
- E = Vitamin C within Corn Diet

Significance denotations:
- †(P ≤ 0.10); *(P ≤ 0.05); **(P < 0.01)
Results
Plasma Vitamin C

- Vitamin C effect ($P = 0.04$)
- Vitamin C within HS ($P = 0.01$)
- Vitamin C within S ($P = 0.02$)
Plasma Total Antioxidant

Linear effect of S, d 90: $P = 0.003$
Linear effect of S, d 143: $P = 0.10$
# Feedlot Performance Data

<table>
<thead>
<tr>
<th>Diet S</th>
<th>Low</th>
<th>Medium</th>
<th>High</th>
<th>SEM</th>
<th>Sig¹,²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diet vitamin C</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Pen (steers)</td>
<td>5 (20)</td>
<td>5 (20)</td>
<td>5 (20)</td>
<td>5 (20)</td>
<td>5 (19)</td>
</tr>
<tr>
<td>Initial BW, kg</td>
<td>342</td>
<td>342</td>
<td>341</td>
<td>343</td>
<td>340</td>
</tr>
<tr>
<td>Intake</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DMI, kg/d³</td>
<td>10.54</td>
<td>10.50</td>
<td>10.82</td>
<td>10.20</td>
<td>9.41</td>
</tr>
<tr>
<td>S intake, g/d³</td>
<td>21.8</td>
<td>22.6</td>
<td>34.6</td>
<td>33.6</td>
<td>53.6</td>
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<tr>
<td>VC intake, g/d³</td>
<td>-</td>
<td>10.5</td>
<td>-</td>
<td>10.3</td>
<td>-</td>
</tr>
<tr>
<td>Live performance⁴</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Final BW, kg</td>
<td>576</td>
<td>568</td>
<td>575</td>
<td>573</td>
<td>545</td>
</tr>
<tr>
<td>ADG, kg/d</td>
<td>1.65</td>
<td>1.65</td>
<td>1.69</td>
<td>1.63</td>
<td>1.43</td>
</tr>
<tr>
<td>G:F</td>
<td>0.16</td>
<td>0.15</td>
<td>0.16</td>
<td>0.15</td>
<td>0.15</td>
</tr>
</tbody>
</table>

¹Contrast Statements: A = vitamin C vs. no vitamin C; B = Linear effect of sulfur; C = vitamin C within high sulfur; D = vitamin C within sulfur; E = vitamin C within corn diet

²†(P ≤ 0.10); *(P ≤ 0.05); **(P < 0.01)

³Month and treatment × month (P < 0.05)

⁴Live performance values based on measured live BW with a 4% pencil shrink applied
## Carcass Data

<table>
<thead>
<tr>
<th>Diet S</th>
<th>Low</th>
<th>Medium</th>
<th>High</th>
<th>SEM</th>
<th>Sig(^1,2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diet vitamin C</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Pen (steers)</td>
<td>5 (20)</td>
<td>5 (19)</td>
<td>5 (20)</td>
<td>5 (19)</td>
<td>5 (18)</td>
</tr>
<tr>
<td>HCW, kg</td>
<td>369</td>
<td>365</td>
<td>371</td>
<td>367</td>
<td>350</td>
</tr>
<tr>
<td>Dressing %</td>
<td>64.0</td>
<td>64.1</td>
<td>64.8</td>
<td>65.0</td>
<td>64.3</td>
</tr>
<tr>
<td>Fat, cm</td>
<td>1.48</td>
<td>1.28</td>
<td>1.28</td>
<td>1.24</td>
<td>1.00</td>
</tr>
<tr>
<td>REA, cm(^2)</td>
<td>80.6</td>
<td>79.9</td>
<td>81.8</td>
<td>81.8</td>
<td>82.1</td>
</tr>
<tr>
<td>KPH</td>
<td>2.08</td>
<td>2.34</td>
<td>2.21</td>
<td>2.43</td>
<td>2.26</td>
</tr>
<tr>
<td>Calculated YG</td>
<td>3.45</td>
<td>3.28</td>
<td>3.26</td>
<td>3.20</td>
<td>2.90</td>
</tr>
<tr>
<td>Marbling score(^3)</td>
<td>511</td>
<td>443</td>
<td>474</td>
<td>438</td>
<td>398</td>
</tr>
<tr>
<td>QG(^4)</td>
<td>3.45</td>
<td>3.15</td>
<td>3.45</td>
<td>3.00</td>
<td>2.75</td>
</tr>
</tbody>
</table>

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\(^2\)†(\(P \leq 0.10\)); *(\(P \leq 0.05\)); **(\(P < 0.01\))

\(^3\)Marbling Scores, Traces: 200, Slight: 300, Small: 400, Modest: 500, Moderate: 600

\(^4\)Quality Grade refers to the plant assigned QG
### Proximate analysis

<table>
<thead>
<tr>
<th>Diet sulfur</th>
<th>Low</th>
<th>Medium</th>
<th>High</th>
<th>SEM</th>
<th>Sig¹,²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diet vitamin C</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Item</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pen (steers)</td>
<td>5(13)</td>
<td>5(13)</td>
<td>5(15)</td>
<td>5(15)</td>
<td>5(14)</td>
</tr>
<tr>
<td>Moisture, %</td>
<td>71.9</td>
<td>72.2</td>
<td>72.1</td>
<td>72.4</td>
<td>73.2</td>
</tr>
<tr>
<td>Fat, %</td>
<td>5.40</td>
<td>5.03</td>
<td>5.48</td>
<td>5.05</td>
<td>3.56</td>
</tr>
<tr>
<td>Protein, %</td>
<td>22.3</td>
<td>22.1</td>
<td>21.5</td>
<td>22.0</td>
<td>22.3</td>
</tr>
</tbody>
</table>

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²†(P ≤ 0.10); *(P ≤ 0.05); **(P < 0.01)
μ-Calpain autolysis

Vitamin C ($P = 0.09$); vitamin C within high S ($P < 0.05$); vitamin C within C medium and high S ($P = 0.03$)

Linear effect of S ($P = 0.107$)

Linear effect of S ($P = 0.03$); vitamin C within high S ($P < 0.09$)

Percentage of Catalytic Subunit Intact

Catalytic Subunits

Day 2 Postmortem

- Low S
- Low S + vitamin C
- Medium S
- Medium S + vitamin C
- High S
- High S + vitamin C

80 kDa
78 kDa
76 kDa
Troponin-T degradation product

Day 2 Postmortem

Peak Height Ratio

Linear effect of S: $P < 0.01$

- Low S
- Low S + vitamin C
- Medium S
- Medium S + vitamin C
- High S
- High S + vitamin C
Protein carbonylation

Peak Height vs. Day 2 Postmortem

Linear effect of S: $P < 0.04$

- Low S
- Low S + vitamin C
- Medium S
- Medium S + vitamin C
- High S
- High S + vitamin C
### TBARS and Iron analysis

<table>
<thead>
<tr>
<th>Diet S</th>
<th>Low</th>
<th>Medium</th>
<th>High</th>
<th>SEM</th>
<th>Sig¹,²</th>
</tr>
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<tbody>
<tr>
<td>Diet vitamin C</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Pen(steers)</td>
<td>5(18)</td>
<td>5(17)</td>
<td>5(19)</td>
<td>5(20)</td>
<td>5(18)</td>
</tr>
<tr>
<td>Fe, mg/kg</td>
<td>12.8</td>
<td>14.7</td>
<td>12.7</td>
<td>13.8</td>
<td>14.0</td>
</tr>
<tr>
<td>Pen(steers)</td>
<td>5(13)</td>
<td>5(13)</td>
<td>5(12)</td>
<td>5(17)</td>
<td>5(12)</td>
</tr>
<tr>
<td>MDA³, µM</td>
<td>0.29</td>
<td>0.33</td>
<td>0.29</td>
<td>0.24</td>
<td>0.27</td>
</tr>
</tbody>
</table>

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²†(P ≤ 0.10); *(P ≤ 0.05); **(P < 0.01)

³Malondialdehyde (lipid peroxidation product)
• Increasing dietary S
  • Decreased the presence of the 76 kDa subunit of the protease µ-calpain, troponin-t degradation, protein carbonylation, & percent fat in rib facings
  • Increased: percent protein of rib facings

• Addition of vitamin C
  • Increased marbling scores of high S cattle
  • Increased fatness of cattle
  • Increased 76 kDa subunit of µ-calpain in all treatments, but specifically noted in high S
  • Increased TBARS and Fe concentration in muscle
Conclusion and Implications

• Adding vitamin C to high S diets may increase both the quality grade and tenderness of the final meat product

• DDGS-based diets are often high in S, feeding vitamin C to cattle may allow producers enjoy the economic benefits of a DDGS-based diet while still maintaining the quality and tenderness expected by consumers
Acknowledgments

- Danielle Pogge, Ph.D. student
- Dr. Steven Lonergan, meat scientist
- Farm crew at ISU Beef Nutrition Farm
- ICPB and IBIC for funding
- RMC for the opportunity to speak with you!
Got Orange Juice?