Use of organic acids for control of Clostridium perfringens in cooked vacuum-packaged ground beef products subjected to substandard cooling procedures

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Abstract
This study determined the ability of Clostridium perfringens spores to germinate and grow after different organic acid treatments in vacuum packaged cooked ground beef subjected to substandard (slow) cooling. Meat samples were inoculated with a three-strain cocktail of C. perfringens spores (ATCC 10388, NCTC 8238, and NCTC 8239), then vacuum-packaged, cooked in a water bath to 167°F internal temperature, and held 20 min. The water bath temperature was then lowered to 130°F, and samples were cooled from 130°F to 45°F over 18 hr. Samples were taken after inoculation, after cooking, and after cooling. In the event of substandard cooling, sodium citrate at 2 or 4.8% or sodium lactate at 4.8% will control C. perfringens growth, with 4.8% sodium citrate showing the best inhibition.

Keywords
Cattlemen's Day, 2002; Kansas Agricultural Experiment Station contribution; no. 02-318-S; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 890; Beef; Clostridium perfringens; Cooked ground beef; Organic acids

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USE OF ORGANIC ACIDS FOR CONTROL OF CLOSTRIDIUM PERFRINGENS IN COOKED VACUUM-PACKAGED GROUND BEEF PRODUCTS SUBJECTED TO SUBSTANDARD COOLING PROCEDURES

J. R. Sabah, T. Harshavardhan,
J. L. Marsden, and D.Y.C. Fung

Summary

This study determined the ability of Clostridium perfringens spores to germinate and grow after different organic acid treatments in vacuum packaged cooked ground beef subjected to substandard (slow) cooling. Meat samples were inoculated with a three-strain cocktail of C. perfringens spores (ATCC 10388, NCTC 8238, and NCTC 8239), then vacuum-packaged, cooked in a water bath to 167°F internal temperature, and held 20 min. The water bath temperature was then lowered to 130°F, and samples were cooled from 130°F to 45°F over 18 hr. Samples were taken after inoculation, after cooking, and after cooling. In the event of substandard cooling, sodium citrate at 2 or 4.8% or sodium lactate at 4.8% will control C. perfringens growth, with 4.8% sodium citrate showing the best inhibition.

(Key Words: Clostridium perfringens, Cooked Ground Beef, Organic Acids.)

Introduction

Between 1983 and 1992, Clostridium perfringens caused 8.94% of all reported food bacterial outbreaks, and 4.77% of total estimated cases of bacterial disease in the United States. Changing lifestyles have increased use of pre-cooked, vacuum-packaged food. In such products, competitive normal microflora, which serve as “spoilage indicators,” are destroyed. Thus, pathogen contamination can be present even though the product looks and smells normal.

Clostridium perfringens is a heat resistant, spore-forming bacteria that can survive the mild heat treatments used for ready-to-eat meats. Such mild heating may even serve to “activate” spores, allowing them to germinate and multiply, especially if the product is cooled slowly after cooking. To prevent this potential health hazard, the USDA requires that the relative growth of C. perfringens should not exceed one log_{10} (a 10× increase) in meat and poultry products. In 1993, the Food and Drug Administration (FDA) recognized that inadequate cooling was a major food safety problem and recommended that all foods should be cooled from 140°F down to about 40°F in 6 hr or less, but did not require changes in the operating performance of refrigerators. Because of the possibility of inadvertently chilling pre-cooked food too slowly, several additives have been examined as a way to protect against foodborne pathogens such as C. perfringens.

Our purpose was to assess the use of certain food grade organic acids and their salts as added protection against C. perfringens growth brought on by an inadequate cooling.

Experimental Procedures

Ground beef from the lean inside round (1/8 inch grind) was obtained from the
Kansas State University Meat Laboratory, and formulated to an industrial recipe; 10% water, 1.5% sodium chloride, and 0.5% trisodium phosphate were added and mixed in a Hobart mixer. Generic sodium citrate at pH 5.6, sodium citrate adjusted to pH 5.0 (Ingredients Solutions Inc., Searsport, Maine), and sodium lactate (Purac, Lincolnshire, Illinois) were used at concentrations of 2% and 4.8%. Sodium acetate and sodium diacetate (Niacet Corporation, Niagara Falls, New York) were used at a 0.25% concentration.

Strains of *C. perfringens* (NCTC 10388, 8238, and 8239) were obtained from the Kansas State University Food Microbiology Laboratory culture collection. An equivalent proportion of spores from each strain were mixed and added to ground beef to get a three-strain cocktail inoculum of 2 log$_{10}$ cfu/g of meat. Samples of prepared ground beef (25 g) were placed in 2 × 3 inch plastic bags, vacuum packaged to a negative pressure of 1000 millibars and heat sealed. The bags were fully submerged and cooked in water at 167°F for 20 min, then chilled from 130°F to 45°F over an 18 hr period; a rate of cooling slower than FDA recommendations.

Samples were taken after inoculation, after cooking, and after the 18-hr cooling period. Fifty ml of 0.1% sterile peptone water was added to each sample and macerated in a Stomacher Lab-blender™ for 2 min. Decimal serial dilutions were prepared in 0.1% peptone water. Total cell counts were obtained by pouring (in duplicate) 1 ml samples into Fung’s Double Tubes using tryptose-sulfite agar without egg yolk. Tubes were incubated at 99°F for 8 to 10 hr before assessment of growth.

**Results and Discussion**

The effects of the different organic acid treatments (mean of two replications) on *C. perfringens* outgrowth are shown in Table 1. Controls showed a 2 log$_{10}$ growth (from 1.61 to 3.87 log$_{10}$ CFU/g) between inoculation and the completion of slow cooling. Similar 1.5 to 2 log$_{10}$ growth was noted in the presence of 0.25% sodium acetate or sodium diacetate. With 2% sodium lactate, *C. perfringens* increased from 1.96 log$_{10}$ to 2.58 log$_{10}$ CFU/g. For all other treatments (sodium citrate at 2% and 4.8% at pH 5.6, and pH 5.0, and sodium lactate at 4.8%) viable *C. perfringens* cells decreased by 0.5 to almost 1 log$_{10}$. Sodium lactate at 4.8% was the most effective growth inhibitor with a 0.96 log$_{10}$ CFU/g reduction.

**Table 1. Log$_{10}$ CFU/g of the Total Clostridium perfringens Cells in Ground Beef**

<table>
<thead>
<tr>
<th>Percent Added</th>
<th>After Inoculation</th>
<th>After Heating</th>
<th>After Slow (18 Hr) Cooling</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium citrate, pH 5.6</td>
<td>2</td>
<td>1.78</td>
<td>2.36</td>
</tr>
<tr>
<td>Sodium citrate, pH 5.6</td>
<td>4.8</td>
<td>1.77</td>
<td>2.29</td>
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<tr>
<td>Sodium citrate, pH 5.0</td>
<td>2</td>
<td>1.84</td>
<td>2.10</td>
</tr>
<tr>
<td>Sodium citrate, pH 5.0</td>
<td>4.8</td>
<td>1.92</td>
<td>2.51</td>
</tr>
<tr>
<td>Sodium lactate</td>
<td>2</td>
<td>1.96</td>
<td>2.39</td>
</tr>
<tr>
<td>Sodium lactate</td>
<td>4.8</td>
<td>2.49</td>
<td>2.31</td>
</tr>
<tr>
<td>Sodium acetate</td>
<td>0.25</td>
<td>1.90</td>
<td>2.43</td>
</tr>
<tr>
<td>Sodium diacetate</td>
<td>0.25</td>
<td>1.96</td>
<td>2.35</td>
</tr>
<tr>
<td>Control</td>
<td>1.61</td>
<td>2.50</td>
<td>3.87</td>
</tr>
</tbody>
</table>