

## THERMAL TREATMENTS FOR INACTIVATION

### INACTIVATION OF *SALMONELLA* IN MANURE-BASED COMPOSTS WITH VARYING C:N RATIOS

(M. C. Erickson, J. Liao, L. Ma, X. Jiang, and M. P. Doyle)

Composting is a process whereby organic matter is decomposed by microorganisms to generate a stable amendment that improves soil quality and fertility. To date, the primary criteria for ensuring the microbiological safety of composts have been adherence to narrowly defined time-temperature conditions. To expand the guidelines whereby inactivation of pathogens could be assured, this study sought to determine if the carbon:nitrogen (C:N) ratio or the presence of ammonium sulfate affects the inactivation of *Salmonella* spp. in cow manure-based compost mixtures. Evaluation of compost conditions on pathogen inactivation was conducted using a bioreactor system. The days to achieve non-detection of *Salmonella* spp. by enrichment culture was used as the endpoint. In addition to pathogen levels, pH and temperature were monitored at 4 locations within the bioreactor. Location within the bioreactor was not a significant variable affecting pathogen inactivation. Compost preparations with an initial C:N ratio of 20:1 required a maximum of 4 days of storage before *Salmonella* was not detected whereas 30:1 and 40:1 C:N preparations required up to 7 days of storage. Both 20:1 and 30:1 C:N preparations were characterized by a decrease in pH to 5.5-5.7 before pH values increased to > 8. In contrast, pH values of 40:1 C:N preparations increased immediately to > 8, generally within the first day of storage. Maximum temperatures encountered in 20:1 C:N preparations for inactivation of pathogens were less than 50°C. Consequently, the cumulative heat exposure required for pathogen inactivation in 20:1 C:N preparations was five-fold less than in 40:1 C:N preparations. Temperatures within preparations supplemented with 0.08% ammonium sulfate were higher than unsupplemented preparations during the first 2 days of storage, however; these higher temperatures did not consistently translate into more rapid rates of pathogen inactivation.

### SURROGATE BACTERIA FOR IN-PLANT CRITICAL CONTROL POINT VALIDATION OF THERMAL INACTIVATION OF *LISTERIA MONOCYTOGENES*

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Heat treatment of meat and poultry is among the most common of processing techniques to assure their microbiological safety and is considered a critical control point in the Hazard Analysis Critical Control Point (HACCP) system. However, it is not feasible in processing plants to validate thermal processes on a periodic basis using pathogenic bacteria. Hence, a suitable non-pathogenic (surrogate) microorganism is needed for process validation and verification. The goal of this research was to validate the relationship of thermal destruction of the surrogate non-pathogenic *Enterococcus* sp. B2354 (formerly known as *Pediococcus* sp. NRRL B-2354 and *Micrococcus freudenteihii*) to that of pathogens of concern in meat products (*Listeria monocytogenes* and *Salmonella*).

Several trials were done to determine the heat resistance of *Enterococcus* sp. B2354, *L. monocytogenes* 101M, and *S. Senftenberg* 775W at four temperatures (58, 62, 65, and 68°C) in ground beef of 4% (lean) and 12% (normal) fat content. In lean ground beef, *L. monocytogenes* was more sensitive to thermal inactivation at 58 and 62°C than *S. Senftenberg*, but slightly more resistant at temperature above 62°C. However, in normal ground beef, *L. monocytogenes* was consistently more heat sensitive than *S. Senftenberg* at all four temperatures tested. Higher fat content protects bacteria from thermal inactivation, especially at temperatures lower than 68°C. D-values for *Enterococcus* sp. B2354 in lean and normal ground beef were 4.5 to 18 and 3.6 to 15 times greater, respectively, than those for the most resistant pathogenic microorganisms (*L. monocytogenes* or *S. Senftenberg* 775W) at all temperatures tested, with the greatest difference in D-values occurring at 58° and 62°C. These results indicate that thermal treatments of ground beef at 58° to 68°C that kill *Enterococcus* sp. B2354 will also kill *Salmonella* and *L. monocytogenes*. Hence, depending on the margin of safety desired, processors could

use this strain of *Enterococcus* sp. B2354 as a surrogate for validation studies of thermal processes in lean and normal ground beef at 58° to 68°C.

In search for a less heat resistant surrogate than *Enterococcus* sp. B2354, the heat resistance of two other potential surrogate microorganisms, *Pediococcus parvulus* HP and *Pediococcus acidilactici* LP, isolated from a commercial meat starter culture, was compared with the three strains under study (*L. monocytogenes* 101M, *S. Senftenberg* 775W, and *Enterococcus* sp. B2354) in broth at 62°C. D-values of *P. parvulus* HP and *P. acidilactici* LP were lower than those of *Enterococcus* sp. B2354 but 4.1 and 2.5 times greater, respectively, than those of the most resistant pathogen (*S. Senftenberg* 775W). Therefore, these two *Pediococcus* strains may serve as alternate surrogates for validation studies when a less heat resistant surrogate is desired; however, studies at additional temperatures are needed with these strains for validation of the entire range of 58° to 68°C.

### **THERMAL TOLERANCE OF ACID-ADAPTED AND UNADAPTED SALMONELLA, ESCHERICHIA COLI O157:H7, AND LISTERIA MONOCYTOGENES IN CANTALOUPE JUICE AND WATERMELON JUICE**

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Outbreaks of foodborne infections associated with the consumption of fresh fruits and vegetables as well as unpasteurized juices contaminated with pathogenic bacteria have been documented. Outbreaks of salmonellosis and *Escherichia coli* O157:H7 infections have been linked to the consumption of cantaloupes. Watermelons have been implicated in outbreaks of salmonellosis and shigellosis. Pathogens known to be contaminants on the surface of melon rinds can be translocated to the edible tissues and juices when melons are cut to prepare for consumption. *Salmonella* can rapidly grow on sliced cantaloupe, watermelon, and honeydew melon, and in cantaloupe juice and watermelon juice. *Escherichia coli* O157:H7 has been reported to grow on cantaloupe and watermelon cubes and *Listeria monocytogenes* can grow in cantaloupe and watermelon pulp. The U.S. Food and Drug Administration has implemented a HACCP program that focuses on minimizing microbiological safety risks that may be associated with fruit and vegetable juices. One of the interventions to eliminate foodborne pathogens is heat treatment. The use of melon juice in blends of non-pasteurized and pasteurized fruit juices offered for sale to the consumer has increased in recent years. To date, research efforts on the microbiological safety of pasteurization processes for fruit juices have concentrated largely on determining *D* values (decimal reduction times) for *Salmonella*, *E. coli* O157:H7, and *L. monocytogenes* in apple juice. We undertook a study to determine the *D* values of these pathogens in cantaloupe juice and watermelon juice as affected by acid adaptation preceding exposure to heat.

*Salmonella enterica* serotype Poona, *Salmonella enterica* serotype Saphra, two strains of *E. coli* O157:H7, and two strains of *L. monocytogenes* were grown in tryptic soy broth (TSB) and TSB supplemented with 1% glucose for 24 h at 37°C. Decimal reduction times (*D* values) of cells suspended in unpasteurized cantaloupe juice and watermelon juice were determined. Acid-adapted cells of *Salmonella* and *E. coli* O157:H7, but not *L. monocytogenes*, had increased thermal tolerance compared to cells that were not acid-adapted. There was no correlation between soluble solids content of the two types of juice and thermal resistance. Growth of *Salmonella* and *E. coli* O157:H7 in cantaloupe juice, watermelon juice, or other acidic milieu, either in preharvest or postharvest environments, may result in cross protection to heat. The pasteurization conditions necessary to achieve elimination of pathogens from these juices would consequently have to be more severe if cells are habituated to acidic environments. Insights from this study provide guidance to developing pasteurization processes to eliminate *Salmonella*, *E. coli* O157:H7, and *L. monocytogenes* in cantaloupe juice and watermelon juice.