

## **Microbial Validation of Biltong Processing to Achieve 5-Log Reduction of *L. monocytogenes* and *E. coli* O157:H7**

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**Introduction:** ‘Biltong’ is a South African style dried beef product that is marinated and dried at moderate heat but still must accommodate USDA-FSIS safety concerns for process validation by demonstrating either negative ingredient pathogen testing and a 2-log process reduction of pathogen, or simply a process achieving a 5-log pathogen reduction.

**Purpose:** Our objective was to achieve a 5-log reduction of *Listeria monocytogenes* and *E. coli* O157:H7 without a heat lethality step and evaluate the individual contribution of each component of the marinade (spice, salt, vinegar) to the overall reduction on inoculated beef.

**Methods:** Beef was cut into small ‘steaks’ (0.75-in x 2-in x 3-in) and inoculated with a 4-serovar mixture of acid-adapted *Listeria monocytogenes* (or acid-adapted *E. coli* O157:H7) on each side and spread using the gloved finger technique. The beef pieces were refrigerated for 30 minutes for the attachment of the bacteria followed by water dip treatment for 30 seconds. Beef pieces were then vacuum-tumbled with either a mixture of spice (1.1% coriander and 0.8% black pepper), 2.2% salt and 4% 100 grain vinegar of total formulation or just with individual marinade components, and marinated for 30 minutes. Beef was then dried in a temperature-controlled humidity oven for 10 days (75°F/23.9°C; 55% RH). Beef pieces were randomly selected and stomached with 100 ml of chilled 1% neutralizing buffered peptone water followed by 10-fold serial dilution with 0.1% BPW for microbial enumeration performed post-marination, and after 2, 4, 6, 8, and 10 days of drying.

**Results:** The combination of spice, salt, and vinegar resulted in a reduction of >5-log and >6-log with subsequent drying period drying of 8 and 10 days, respectively. The greatest reduction of *L. monocytogenes* from the individual components of the marinade was exhibited by vinegar(>5-log) followed by salt (4.7-log) and spice (4.1 log). The conditions of drying, without any added ingredients, rendered a 4.2-log reduction over a period of 10 days and water activity of 0.79. All trials were performed in duplicate replication (triplicate samples) and repeated measures one-way ANOVA to determine significant differences ( $p < 0.05$ ).

**Significance:** This is the first published work achieving >5-log reduction of *L. monocytogenes* and *E. coli* O157:H7 during the processing of biltong, thereby validating this process for USDA-FSIS approval that is sought by many upscale food stores to ensure product safety.

**Keywords:** Biltong, *Listeria monocytogenes*, *E. coli* O157:H7, 5-log reduction, process validation.