Validation of Ground-and-Formed Beef Jerky Processing Lethality Using Commercial Lactic Acid Bacteria Starter Cultures

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Introduction:

•Popularity of jerky products has increased substantially due to the low-carb diet fad, experiencing a 147% increase in sales between 1997 and 2002 (7).
•Outbreaks of salmonellosis, linked to beef jerky, have been occurring for the past 40 years and *E. coli* O157:H7 outbreaks have been linked to both beef and venison jerky (1).
•Inherent characteristics of the jerky manufacturing process lead to the increased possibility of producing an unsafe product:
- •Low-temperature drying for long periods of time decreases the degree of pathogen destruction,
- •Evaporative cooling on the surface of jerky strips decreases the temperature to which any contaminating organisms are exposed,
- •Heat-resistance of *Salmonella* serovars and *E. coli* O157:H7 increases as the water activity (a_w) of the product decreases.

•Previous research has not clearly defined the processing conditions required to achieve adequate pathogen lethality in jerky manufacture (2,3,4,5), while conceding that recommended processes often yield a poor quality product (9).
•Processors are therefore left with little useful information on how to safely manufacture beef jerky, especially the ground-and-formed product.
•Ideally validation of a process would be achieved via a challenge study but that is not feasible for most small ground-and-formed beef jerky processors.
•Using lactic acid bacteria (LAB) as a pathogen surrogate is a safe way for processors to perform a "challenge" study under their own unique processing conditions and evaluate the safety of their process.

Objective:

To validate the use of a commercial LAB pathogen-surrogate for evaluating the lethality of ground-and-formed beef jerky processes. It is important to note that it WAS NOT the goal of the study to validate any of the processes used but rather to expose the LABs to a variety of processing conditions to ensure the LABs exhibited greater, but comparable, heat resistance to *Salmonella* spp. and *E. coli* O157:H7.

Materials and Methods:

Inoculum Preparation:
•Ten-strain cocktail containing five strains each of *Salmonella* serovars and *E. coli* O157:H7 re-suspended in Butterfield's phosphate diluent (BPD).
•0.5g of *Pediococcus* spp. or *Pediococcus acidilactici* re-suspended in 9mL BPD.

Inoculation of Jerky Batter:
•53g of lean ground beef placed in 3.785 L Ziploc bags and seasoned by hand mixing with "Colorado" seasoning (Excalibur Seasoning Company, Pekin, IL) according to manufacturer's directions.
•8mL of either LAB culture or pathogen cocktail added to the seasoned ground beef in the Ziploc bag and again mixed by hand.

Formation of Jerky Strips:
•Inoculated jerky batter was placed on a sterilized rimmed tray and rolled to a depth of 0.64 cm using a sterilized Nalgene bottle.
•Strips were cut into 2.5 cm x 12.5 cm strips with a sterile pizza cutter - yielding 18, 2.5 cm x 12.5 cm x 0.64 cm strips per tray.
•This process replicated methods used by small meat processors to manufacture larger batches (22.5-45.5. kg) of ground-and-formed jerky (personal observation).

N=100		*Pediococcus* spp. Death (log CFU)	
		< 4	≥ 4
E. coli O157:H7 Death (log CFU)	< 5	43	0
	≥ 5	31	26

N=100		*Pediococcus* spp. Death (log CFU)	
		< 4	≥ 4
Salmonella spp. Death (log CFU)	< 5	57	1
	≥ 5	17	25

N=100		*P. acidilactici* Death (log CFU)	
		< 4	≥ 4
E. coli O157:H7 Death (log CFU)	< 5	43	2
	≥ 5	26	29

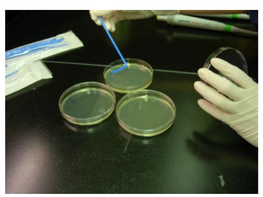
N=100		*P. acidilactici* Death (log CFU)	
		< 4	≥ 4
Salmonella spp. Death (log CFU)	< 5	55	4
	≥ 5	14	27

Figure 1. Matrices comparing jerky process lethality (log CFU) against *E. coli* O157:H7 or *Salmonella* spp. and *Pediococcus* spp. or *Pediococcus acidilactici*. Combined results from all six jerky processes are presented. Each number represents a sample pair formed by comparing the lethality against a pathogen and an LAB determined from two separate jerky strips at a given sampling time.

Jerky Processing Conditions:
•Once cut, strips were either:
- Removed from the tray and placed on racks for processing in a small commercial dehydrator (Pragotrade model TS160, Cabela's Inc., Sidney, NE), or
- Trays were covered in foil and placed in coolers with ice packs for transport to the Alkar-RapidPak Research and Technology Center for processing in a commercial smokehouse (Model 2000, Alkar, Lodi, WI).

•In total 6 separate processes were evaluated (Table 1).

Enumeration of Surviving Cells:
•Two jerky strips were removed from the dehydrator or smokehouse at designated sampling times throughout each process.
•The strips were stomached separately for 2 minutes at medium speed (Stomacher 400 Circulator lab blender, Seward, Worthington, UK) in 99mL of BPD, diluted and subsequently direct-plated on BHIA.
•Plates were incubated for 1 hr and then overlaid with XLD agar (*E. coli* O157:H7 and *Salmonella* spp)., or MRS agar (LABs) to allow for recovery of injured cells.
•Samples were incubated at 35ºC for 24 hrs (XLD) or 48 hrs (MRS), and log CFU calculated for each sample. A value of 0.5 CFU was assigned when the lowest dilution had no countable colonies.



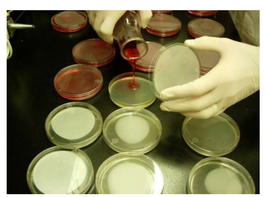


Table 1. Summary of Jerky Processes

Process	Step Time (min)	Set Dry Bulb Temp. ºC (ºF)	Set Wet Bulb Temp. ºC (ºF)	Actual Dry Bulb Temp. Range ºC (ºF)	Actual Wet Bulb Temp. Range ºC (ºF)
1	420	68.3 (155)	NC[a]	49.4-66.1 (121-151)	13.9-38.3 (57-101)
2	30	76.7 (170)	60.0 (140)	57.8-77.2 (136-171)	51.7-61.1 (125-142)
	120	54.5 (130)	NC	52.8-57.8 (127-136)	28.9-37.2 (84-99)
	90	76.7 (170)	NC	76.1-77.8 (169-172)	48.3-55.6 (119-132)
3	30	76.7 (170)	NC	76.1-77.8 (169-172)	30.0-41.7 (86-107)
	15	76.7 (170)	71.7 (161)	65.6-78.3 (150-173)	33.9-56.7 (93-134)
	130	76.7 (170)	NC	76.7-80.6 (170-177)	ND[b]
4	90	57.2 (135)	51.7 (125)	55.6-58.9 (132-138)	47.8-52.2 (118-126)
	150	85.0 (185)	NC	83.9-88.9 (183-192)	30.6-55.6 (87-132)
5	90	54.5 (130)	NC	53.3-57.8 (128-136)	31.7-37.8 (89-100)
	60	60.0 (140)	21.1 (70)	58.9-62.8 (138-145)	39.5-42.2 (103-108)
	60	65.6 (150)	26.7 (80)	64.5-67.8 (148-154)	44.5-47.2 (112-117)
	60	70.1 (160)	32.2 (90)	70.6-76.1 (159-169)	50.0-51.7 (122-125)
	60	76.7 (170)	NC	75.6-81.1 (168-178)	53.3-56.1 (128-133)
6	75	51.7 (125)	NC	51.1-52.2 (124-126)	ND
	30	51.7 (125)	NC	50.6-51.7 (123-125)	ND
	75	65.6 (150)	NC	64.5-66.1 (148-151)	ND
	30	85.0 (185)	NC	84.5-87.8 (184-190)	ND

[a] NC, not controlled
[b] ND, not determined

Results and Discussion:

• USDA guidelines require a 5.0 log CFU reduction in *Salmonella* spp. in dried meat products; although not required, a 5.0 log CFU reduction in *E. coli* O157:H7 is also recommended in dried meat products (6,8).
•A reduction in LAB levels of ≥ 4.0 log CFU was found to be a consistent predictor of ≥ 5.0 log CFU reduction in both *Salmonella* spp. and *E. coli* O157:H7.
•When a ≥ 4.0 log CFU lethality against LAB was achieved in a sample, adequate (≥ 5.0 log CFU) lethality against pathogens was achieved in the following percentages of samples (Green values in Figure 1).
- **•*E. coli* O157:H7 ≥ 5.0 log lethality:**
  - **•*Pediococcus* spp. – 100% (n=26)**
  - **•*Pediococcus acidilactici* – 94% (n=32)**
- **•*Salmonella* spp. ≥ 5.0 log lethality:**
  - **•*Pediococcus* spp. – 96% (n=26)**
  - **•*Pediococcus acidilactici* – 87% (n=31)**

•Across all combinations, **instances in which pathogen reduction was < 5.0 log CFU and LAB reduction was ≥ 4.0 log CFU only occurred in 3% of samples (n=205). (Red values in Figure 1)**
•Jerky process schedules resulted in various levels of lethality. Examples of a sub-lethal and lethal process are shown in Figures 2 and 3, respectively.
•Further research reexamined processes 2-6 with addition of smoke during the drying process and also included a second seasoning ("Barbeque", Excalibur Seasoning Company, Pekin, IL). These samples were direct-plated on MEMB agar for the recovery of *Salmonella* spp. and *E. coli* O157:H7 and the MRS overlay method described previously for the recovery of LABs. This work resulted in the following findings:
- •Lethality against both pathogens and LABs was affected by seasoning mix ($p<0.004$),
- •Adding smoke to a process increased lethality ($p<0.0005$), in part due to increased smokehouse temperatures,
- •Initial wet-bulb temperature is critical in determining lethality against pathogens, the higher the better.

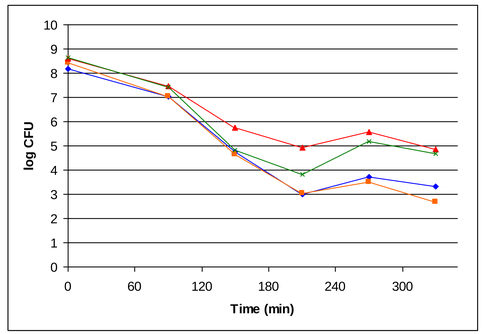


Figure 2. LAB and pathogen lethality throughout process 5.

Key: ♦ *Salmonella* serovars, ■ *E. coli* O157:H7, ▲ *Pediococcus* spp., and x *P. acidilactici*.

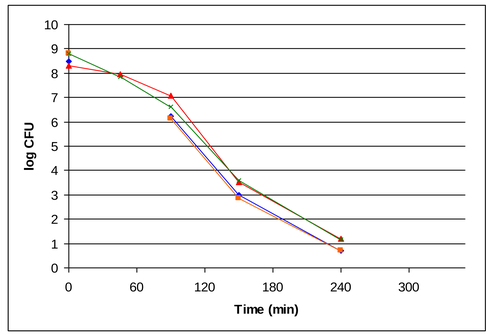


Figure 3. LAB and pathogen lethality throughout process 4.

Key: ♦ *Salmonella* serovars, ■ *E. coli* O157:H7, ▲ *Pediococcus* spp., and x *P. acidilactici*.

Conclusions:

LABs can be used as pathogen surrogates to evaluate process lethality in the manufacture of ground-and formed beef jerky. Achieving a ≥ 4.0 log CFU reduction in a *Pediococcus* spp. population corresponded with a ≥ 5.0 log CFU reduction in pathogens in 96-100% of samples from the processes evaluated. Small meat processors wishing to evaluate the safety of their manufacturing process for ground-and-formed jerky can use readily available LABs and avoid the expense and hazard of challenge studies. Most importantly, this method of process evaluation allows for variation in finished product quality and ultimately maintains the artisanal quality of jerky produced by small and very small meat processors.

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