Survival of *Listeria monocytogenes* in Commercial Cheese Brines

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ABSTRACT

The survival of *Listeria monocytogenes* was determined in commercial cheese brines collected from cheese factories in Wisconsin and northern Illinois. Survival of *L. monocytogenes* inoculated into commercial cheese brines ranged from <7 d to over 259 d. Survival did not correlate with pH, salt content, nitrogen content, mineral content, or inherent microbial populations but was negatively associated with addition of sodium hypochlorite at the dairy plant. The *L. monocytogenes* generally survived longer in brines held at 4°C than at 12°C. Sodium hypochlorite or hydrogen peroxide inactivated *L. monocytogenes* when added to commerical brines in the lab at 10 to 100 ppm or 0.001% to 0.02%, respectively. Addition of 1% potassium sorbate or 1% sodium benzoate also decreased survival of *L. monocytogenes*. Laboratory filtration of commercial brines had a negative effect on survival in one of three brines tested. The *L. monocytogenes* did not grow during incubation in any of the commercial brine samples tested.
(**Key words:** *Listeria monocytogenes*, sodium chloride, cheese brine, survival)

Abbreviation key: BHI = brain heart infusion, **TSB** = trypticase soy broth.

INTRODUCTION

Salting of cheese by immersion in brine is a common industry practice. Several cheese varieties are brine salted, including pasta filata types (mozzarella, provolone, salami, and giganti), brick, Hispanic, and feta. Salt contributes to the flavor in cheeses, as well as creating unfavorable conditions for growth of some microbes by lowering water activity (7). The salt content of brines is maintained at 18 to 24% for most varieties and at 5 to 10% for feta. Brine temperatures are generally maintained at 4 to 10°C.

Cheese brining systems vary in design. In static systems, cheese is immersed in brine-filled tanks. In raceway systems, cheese is conveyed slowly through a brine-filled trough. Salt may be added to the tank or via brine circulation from a brine supply. Either system may include filters or membrane units for removal of particulate material or microorganisms. Periodically, brine may be pasteurized to reduce the microbial load.

Listeria monocytogenes is a halotolerant, Gram-positive, facultatively anaerobic, nonsporeforming rod. The organism is capable of growth at temperatures as low as 1.1°C (18), water activities as low as 0.90 to 0.92 (27), and pH as low as 4.4 (36). The *L. monocytogenes* has been found in a variety of dairy products (13) and has been the cause of outbreaks of listeriosis involving dairy products (11) and other foods.

The *L. monocytogenes* is a ubiquitous organism in the environment with many possible modes of entry into dairy processing facilities. Various studies have reported *L. monocytogenes* in 1.4 to 9.3% of environmental sites tested within dairy processing plants (3, 4, 5, 12, 17, 19, 31, 38). *Listeria monocytogenes* has been isolated from raw milk. In addition, *Listeria innocua* has been isolated from brine solutions in one dairy plant (17). Most evidence suggests that HTST pasteurization of raw milk inactivates *L. monocytogenes* (30).

The *L. monocytogenes* is found more commonly in wet areas of dairy plants, such as floor drains, conveyers, floors, and equipment with condensate (7, 26). The organism has been shown to attach to stainless steel surfaces at different pH and temperature levels in nutrient media (15). Brining system environments are humid, and condensation is routinely present on product, equipment, and building surfaces, posing a risk of cheese contamination with *L. monocytogenes*.

An earlier study found that significant numbers (2.6 $\log_{10}$ cfu/ml) of *L. monocytogenes* migrated from inoculated feta cheese into 12% salt brine over 24 h (28). When the cheese was transferred into 6% salt brine at 22°C for ripening, *L. monocytogenes* initially grew in the brine to populations similar to those in the cheese. During extended incubation of the 6% salt brine, *L. monocytogenes* populations decreased, but the pathogen was still detected after 90 d. In another study (32), *L.*

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monocytogenes migrated from inoculated brick cheese into 22% NaCl brine and was detected in 8 to 50% of brine samples after 5 d at 10°C.

Evidence indicates a possible risk that cheese brines in dairy plants could become contaminated with *L. monocytogenes*, either from environmental sources or by leaching of the organism from cheese contaminated with the pathogen (33). The behavior of *L. monocytogenes* in commercial cheese brine systems is not well characterized. Concerns include the possibility of survival of *L. monocytogenes* for extended periods of time in brines containing high salt levels or its numbers increasing in brines with lower salt concentrations, such as those used for feta brines. The objective of this study was to determine the potential for brine as a reservoir for *L. monocytogenes* by studying survival of the organism in commercial cheese brines.

MATERIALS AND METHODS

Cultures

Listeria monocytogenes strains Scott A, California, Ohio, V7, 2811M (isolated from Mozzarella cheese), and 87189 (isolated from Mozzarella cheese brines) were used in this study. All strains were maintained on brain heart infusion (**BHI**) agar (Difco, Detroit, MI) slants at 4°C. Prior to inoculation into commercial brines or salt solutions, strains were incubated for 18 to 20 h at 37°C in BHI broth, washed twice with 67 m*M* sodium phosphate buffer (pH 6.6), and resuspended in the same buffer.

Collection of Commercial Brine Samples

Thirty-eight commercial brine samples were obtained from 14 cheese plants in Wisconsin and northern Illinois. Brines were kept refrigerated during transport and during storage at the Food Research Institute, Madison, WI.

Information on the commercial brines is presented in Table 1. Twenty-six of the brines were from systems used to salt pasta filata varieties, including mozzarella, string, provolone, fresh salami, and giganti cheeses. One brine each was used to salt romano and parmesan. Seven brines were from systems used to salt brick or brick and Hispanic-style cheese. Three were from feta brine systems. One (F1) was obtained from a fresh brine storage tank and had not yet come into contact with cheese. Both raceway and static tank system brines were represented, as was a variety of filtration equipment.

Analysis and Microbiology of Commercial Brines

Brine pH was determined directly (Research Analyzer model 399A; Orion, Cambridge, MA). Sodium chloride content was determined using the Mohr method of chloride titration (40). Levels of hypochlorite present in brines were determined by potassium iodide titration (39). Brines were analyzed for total nitrogen and minerals, including phosphorous, potassium, calcium, magnesium, sulfur, zinc, boron, manganese, iron, copper, aluminum, and sodium, at the University of Wisconsin Extension Soil and Plant Analysis Laboratory.

Brines were diluted in 67 m*M* sodium phosphate buffer (pH 6.6) and 0.1 ml was spread-plated on plate count agar (Difco). Plates were incubated for 2 d at 30°C to determine aerobic plate counts or 10 d at 4°C to determine psychrotrophic plate counts. Serial dilutions of brines were also plated on YM agar (Difco; pH 3.5), which was incubated at 25°C for 3 to 5 d to determine yeast and mold counts. All microbiological and chemical analyses were conducted on duplicate samples of each commmercial brine. The variation in microbiological counts between samples was ≤20%.

Inoculation of *L. monocytogenes* into Commercial Cheese Brines

Each commercial brine was inoculated with a mixture of *L. monocytogenes* strains Scott A, California, Ohio, V7, 87189, and 2811M in equal concentrations (total inoculum 10^4 to 10^5 cfu/ml). Brines were incubated at 4 or 12°C, and *L. monocytogenes* populations were periodically determined by plating on modified oxford agar plates, which were incubated 2 to 3 d at 37°C. It was not feasible to specifically detect *L. monocytogenes* (e.g., injured cells) on nonselective media because the brines contained a variety of different microorganisms.

Different levels of potassium sorbate (Sigma, St. Louis, MO), sodium benzoate (Mallinckrodt, Paris, KY), sodium hypochlorite (5.25% in bleach), or hydrogen peroxide (Aldrich, Milwaukee, WI) were added to commercial brines B3 (brick cheese) and M9 (mozzarella or string cheese). These two brines were chosen to represent brines from different varieties of cheeses that permitted survival of *L. monocytogenes*. Brines were inoculated with a mixture of the six strains described above and incubated at 4°C, and *L. monocytogenes* survival was monitored by plating as described above. All combinations of variables were tested in duplicate.

Filtration and Inoculation of Commercial Brines

Three commercial brines (M1, B2, and S5) from plants that did not use UF systems were prepared as

TABLE 1. Source, composition of, and survival of *Listeria monocytogenes* in commercial brine samples.[1]

Sample	Plant code	Cheese variety	NaCl (%)	pH	APC[2]	Psychro- trophs	Yeast or mold	System type	Filter types	Sampling point	LM survival[3] 4°C	LM survival[3] 12°C
M1	A	Mozzarella	23.3	4.9	640	<10	<10	Raceway	Coarse	Inlet	189	139
M2	A	Mozzarella	23.8	4.9	380	<10	<10	Raceway	Coarse	Middle of tank	>259	75
M3	A	Mozzarella	24.2	4.9	310	<10	<10	Raceway	Coarse	Outlet	189	75
M4	A	Mozzarella	24.7	4.9	280	<10	<10	Raceway	Coarse	Surge tank	189	115
M5	B	Mozzarella	23.5	5.0	90	<10	<10	Static tank	Coarse/bag	Inlet-tank #1	37	37
M6	B	Mozzarella	23.5	5.0	90	<10	<10	Static tank	Coarse/bag	Outlet-tank #1	139	37
M7	B	Mozzarella	24.1	5.0	140	10	<10	Static tank	Coarse/bag	Inlet-tank #2	37	37
M8	B	Mozzarella	23.5	5.0	100	10	<10	Static tank	Coarse/bag	Outlet-tank #2	37	37
M9	C	Mozz/string	22.5	5.0	1300	<10	<10	Raceway	Coarse/UF	UF outlet	139	139
M10	C	Mozz/string	22.0	5.0	1300	<10	<10	Raceway	Coarse/UF	Middle of tank	>209	139
M11	D	Mozzarella	20.5	5.2	1500	<10	20	Raceway[4]	Coarse/UF	Middle of tank	>215	63
M12	E	Mozzarella	21.3	5.2	25,000	10	80	Raceway	Coarse/in-line	Middle of tank	150	>215
M13	E	Mozzarella	20.5	5.2	33,000	<10	90	Static tank	Coarse	Middle of tank	>215	150
M14	F	Mozzarella	18.9	5.2	540	<10	10	Raceway[4]	Coarse/UF	Middle-tank #1	150	150
M15	F	Mozzarella	16.9	5.2	170	<10	10	Raceway[4]	Coarse/UF	Middle-tank #2	>215	150
M16	F	Mozzarella	18.4	5.2	150	<10	<10	Raceway[4]	Coarse/UF	UF outlet	>215	150
M17	G	Mozzarella	24.6	5.2	170	<10	<10	Raceway	Coarse/UF	Brine silo	62	62
M18	G	Mozzarella	22.4	5.3	2200	<10	15	Raceway	Coarse/UF	Middle of tank	>224	>224
M19	D	Mozzarella	22.0	5.3	2000	<10	10	Raceway[4]	Coarse/UF	Near UF outlet	>215	90
M20	H	Mozzarella	19.7	5.3	29,000	<10	860	Raceway	Coarse/in-line	Middle of tank	>215	>215
M21	H	Mozzarella	19.8	5.3	32,000	<10	560	Raceway	Coarse/in-line	Brine silo	>215	150
M22	I	Mozzarella	21.4	5.4	1000	<10	<10	Raceway[5]	Coarse/UF	Middle of tank	17	21
M23	I	Mozzarella	21.2	5.4	<10	<10	<10	Raceway[5]	Coarse/UF	UF outlet	7	<7
S1	C	Provolone	23.1	5.0	3600	<10	40	Raceway	Coarse/UF	Middle of tank	>209	139
S2	J	Fresh salami	16.8	5.0	18,000	<10	23,000	Static tank[6]	Skimmed	Middle of tank	150	81
S3	J	Giganti	20.6	4.8	32,000	<10	110	Static tank[6]	Skimmed	Middle of tank	150	108
S4	J	Romano	15.8	5.1	29,000	<10	240	Static tank[6]	Skimmed	Middle of tank	108	150
S5	J	Parmesan	18.5	4.9	9500	<10	11,000	Static tank[6]	Skimmed	Middle of tank	150	150
B1	K	Brick	23.0	5.3	13,000	<10	<10	Raceway[4]	Coarse	Outlet-tank #1	>224	150
B2	K	Brick	22.6	5.3	10,000	<10	<10	Raceway[4]	Coarse	Inlet-tank #1	>224	150
B3	K	Brick	22.2	5.3	7000	<10	15	Raceway[4]	Coarse	Inlet-tank #2	>224	150
B4	K	Brick	21.9	5.3	12,000	<10	<10	Raceway[4]	Coarse	Outlet-tank #2	>224	150
H1	L	Brick/Hispanic	24.1	5.4	15	<10	<10	Static tank[7]	Coarse/bag	Middle-tank #2	<7	<7
H2	L	Brick/Hispanic	23.9	5.8	15	<10	<10	Static tank[7]	Coarse/bag	Middle-tank #1	<7	<7
H3	L	Brick/Hispanic	23.5	6.1	360	<10	<10	Static tank[7]	Coarse/bag	Middle-tank #3	<7	<7
F1	M	Feta	6.5	6.8	350	<10	<10	Wet pack	None	Fresh brine tank	118	118
F2	N	Feta	10.0	4.9	13,000	<10	9200	Raceway	Coarse/UF	Middle of tank	118	56
F3	N	Feta	5.6	4.4	3000	<10	1900	Wet pack	None	Bucket	34	13

[1]Initial *L. monocytogenes* inoculum was 2×10^4 to 2×10^5 cfu/ml.

[2]APC = Aerobic plate count.

[3]Day of incubation when *L. monocytogenes* (LM) was last detected.

[4]Glycerol cooling plate used.

[5]Plant reported adding sodium hypochlorite to brines to control yeast and mold; no hypochlorite detected by titration.

[6]Concrete tanks.

[7]Plant reported adding sodium hypochlorite to brines to control yeast and mold; 2 to 5 ppm of hypochlorite detected by titration.

follows: 1) filtered through a coarse filter (Whatman #40 ashless filter, which removes >8-μm particles), 2) filtered through a coarse filter and 0.45-μm syringe filter, and 3) filtered through a coarse filter, 0.45-μm syringe filter, and 0.2-μm syringe filter. Filtered brines were inoculated with a mixture of the six strains described above and incubated at 4°C, and *L. monocytogenes* survival was determined as described above.

Leaching of Sodium Hypochlorite from Aqueous NaCl Solutions into Mozzarella or Brick Cheese

Mozzarella and brick cheeses purchased at a local grocery store were cut into cubes approximately 2.5 cm^3 in size. Cubes were soaked at 4°C in aqueous, 20% NaCl solutions containing 20, 100, or 1000 ppm (milligrams per kilogram) of sodium hypochlorite. Periodically, cubes were removed, and both the outer 3 mm and the inner portions of each cube were tested for the presence of hypochlorite by potassium iodide titration.

RESULTS

Analysis and Microbiology of Commercial Brines

Results of compositional analyses and microbiological determinations of the commercial brines are listed in Table 1. Brine pH ranged from 4.4 to 6.8. Percentage of NaCl ranged from 5.6% to 24.7%. Brines used to salt feta cheese had the lowest NaCl content. Hypochlorite was detected at 2 to 5 ppm in three brines (H1 to H3) from the same plant (plant L). Sodium hypochlorite at an unknown concentration was added to these brines at the cheese plant, as well as to brines M22 and M23 from plant I, to help control microbial populations.

Aerobic plate counts varied widely (<10 to 33,000 cfu/ml) among plants but were relatively consistent in brines from the same plant. Psychrotrophs were detected in low concentrations (10 cfu/ml) in only three brines. Yeast and mold counts ranged from <10 to 23,000 cfu/ml. Brines with high yeast and mold counts generally also had relatively high aerobic plate counts.

Total nitrogen levels ranged from 2.5 ppm (fresh feta brine F3) to 1600 ppm and were relatively similar in brines from the same plants (data not shown). Most brines had total nitrogen levels in the range of 100 to 400 ppm.

Except for brine F3, the mineral content of brines ranged as follows (data not shown in table): sodium, 22,000 to 89,000 ppm; phosphorous, 162 to 1026 ppm; potassium, 201 to 1173 ppm; calcium, 277 to 1993 ppm; magnesium, 27 to 168 ppm; and sulfur, 28 to 201 ppm. Boron, manganese, copper, and aluminum were below detectable levels in all brines. Iron was detected only in brine S4 at 3.4 ppm. Zinc was detected only in brines S2 to S5 and F1 to F2 and ranged from 0.4 to 9.0 ppm in those brines.

Survival of *L. monocytogenes* in Commercial Brines

Table 1 indicates the time that *L. monocytogenes* was detected by direct plating (≥10 cfu/ml) in inoculated commercial brines stored at 4 or 12°C. The *L. monocytogenes* was still detected at the last sampling time (up to 259 d) in several brines. The *L. monocytogenes* populations in one brine used to salt provolone (S1) and four brines (B1 to B4) used to salt brick cheeses decreased only ca. one log cfu/ml during incubation at 4°C for 209 or 224 d, respectively (data not shown). In general, *L. monocytogenes* survived longer in brines stored at 4°C than at 12°C. No correlations were noted between the length of *L. monocytogenes* survival and microbial populations, mineral content, or nitrogen content. In brine F1, a feta brine with the lowest pH (4.4) and salt content (5.6%) of the brines studied, *L. monocytogenes* was detected for only 34 d at 4°C and 13 d at 12°C. In most brines, the decrease in *L. monocytogenes* was relatively linear, rather than showing a lag or presenting as an abrupt decrease followed by a leveling-off (data not shown).

A strong association existed between sodium hypochlorite addition and *L. monocytogenes* survival. In three brines (H1 to H3) with detectable hypochlorite levels, *L. monocytogenes* was not detected at d 7 of incubation. In two brines (M22 and M23), to which sodium hypochlorite was added at unknown levels but in which no hypochlorite was detected, *L. monocytogenes* was detected up to 21 d. Hypochlorite was reported to have been added to these brines at the cheese plant, but at the time of analysis, its level was below the lower limit of detection (~2 ppm) by the method used.

Addition of sodium hypochlorite to commercial brines B3 and M9 in the laboratory significantly decreased *L. monocytogenes* survival when compared with controls (Figures 1A and 1B). In brine B3, *L. monocytogenes* survival was strongly inhibited by 50 to 100 ppm sodium hypochlorite but was not affected by 10 to 20 ppm. In brine M9, 10 ppm of sodium hypochlorite strongly inhibited *L. monocytogenes* survival, whereas 5 ppm caused some inhibition, and ≤2 ppm caused no noticeable inhibition.

Listeria monocytogenes survival was also inhibited by ≥0.02% hydrogen peroxide in brine B3 but was not affected by lower levels (Figures 2A and 2B). In brine M9, moderate inhibition was observed by 0.001% hydrogen peroxide, and strong inhibition occurred with ≥0.005% hydrogen peroxide. Addition of 1% potassium

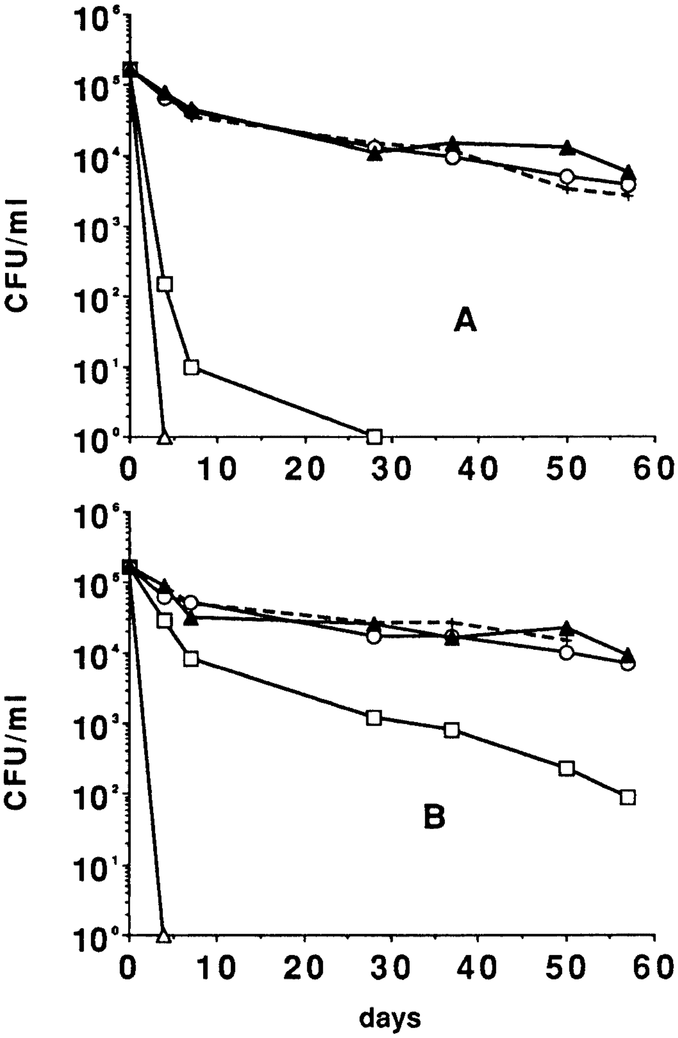

Figure 1. (A) Survival of *Listeria monocytogenes* at 4°C in commercial brine B3 with 0 (+), 10 (▲), 20 (○), 50 (□), or 100 (△) ppm added sodium hypochlorite; (B) survival of *Listeria monocytogenes* in brine M9 with 0 (+), 1 (▲), 2 (○), 5 (□), or 10 (△) ppm added sodium hypochlorite. The points are averages of duplicate samples.

sorbate (Figures 3A and 3B) or 1% sodium benzoate (Figures 4A and 4B) slightly decreased survival of *L. monocytogenes* when added to brines B3 and M9.

L. monocytogenes Survival in Filtered Brines

Filtering brines B2 and S5 through coarse and 0.45-μm filters before inoculation did not strongly affect survival of *L. monocytogenes* at 4°C (Figures 5B and 5C). However, survival of *L. monocytogenes* was decreased in brine M1 filtered through a 0.2-μm filter when compared with survival in unfiltered brine (Figure 5A).

Leaching of Sodium Hypochlorite from Aqueous NaCl Solutions into Mozzarella or Brick Cheese

Because hypochlorite strongly inactivated *L. monocytogenes*, we investigated whether it would migrate from a brine solution into cheese. After 4 d in 1000 ppm sodium hypochlorite, the outer 3 mm of mozzarella samples contained approximately 100 ppm sodium hypochlorite, and the outer 3 mm of brick samples contained approximately 40 ppm sodium hypochlorite. No sodium hypochlorite was detected in either cheese soaked in 50 ppm or 100 ppm or in the inner portion of any cheese cubes tested.

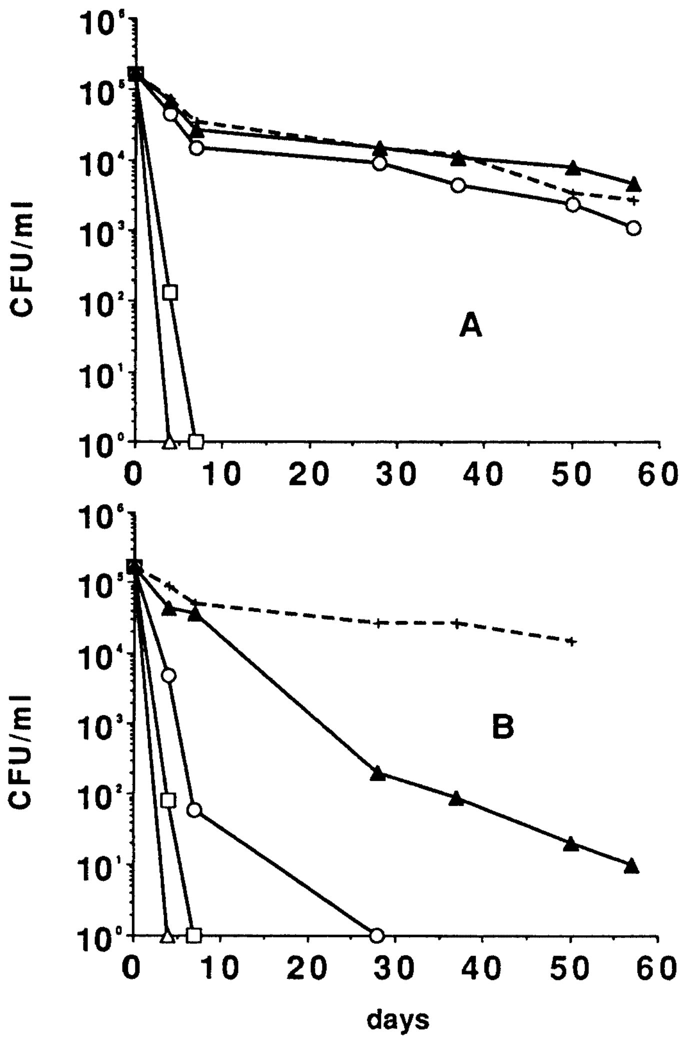

Figure 2. (A) Survival of *Listeria monocytogenes* at 4°C in commercial brine B3 with 0% (+), 0.005% (▲), 0.01% (○), 0.02% (□), or 0.05% (△) added hydrogen peroxide; (B) survival of *Listeria monocytogenes* in brine M9 with 0% (+), 0.001% (▲), 0.005% (○), 0.01% (□), or 0.02% (△) added hydrogen peroxide. The points are averages of duplicate samples.

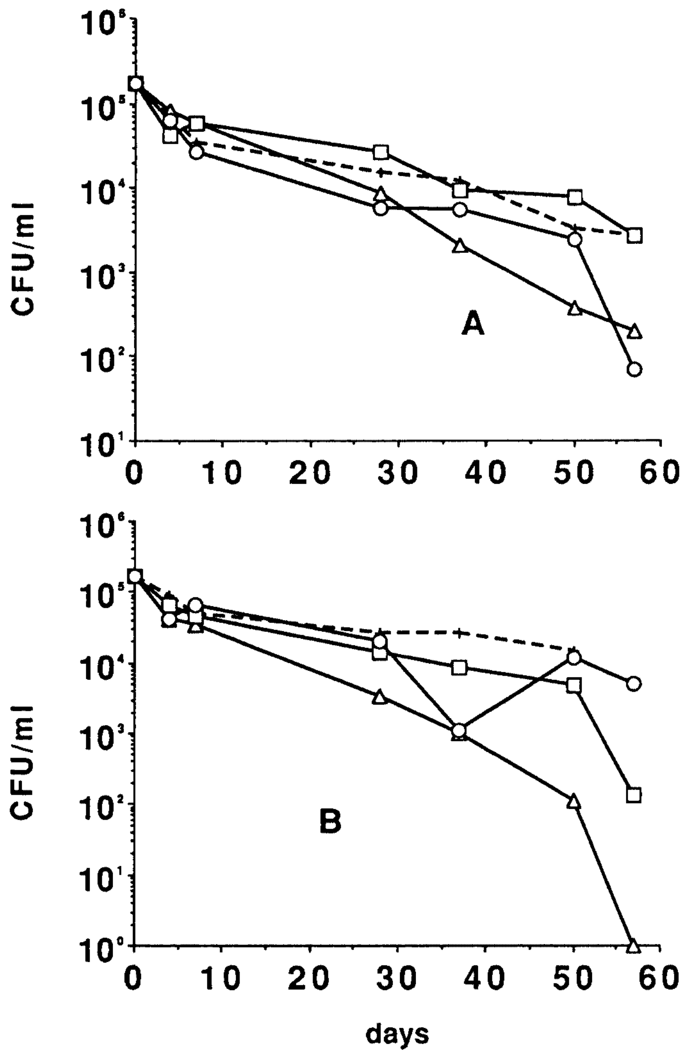

Figure 3. Survival of *Listeria monocytogenes* in commercial brines B3 (A) or M9 (B) with 0% (+), 0.1% (○), 0.5% (□), or 1% (△) added potassium sorbate at 4°C. The points are averages of duplicate samples.

DISCUSSION

Listeria monocytogenes is remarkably salt tolerant and could survive in brining operations in cheese plants. Maximum salt tolerance of *L. monocytogenes* generally occurs at neutral pH (1). Temperature also affects survival of *L. monocytogenes* in the presence of NaCl (14), and greater survival was observed at 4°C than at 22 or 37°C in trypticase soy broth (**TSB**) containing salt. The *L. monocytogenes* can grow in media containing 10% NaCl at 30 and 35°C with fastest growth at neutral pH (22). In another study, survival of *L. monocytogenes* was greater at 10 and 25°C than at 35°C in TSB containing salt (35). In TSB with 25.5% NaCl, *L. monocytogenes* survived 5 d at 37°C, 32 d at 22°C, and more than 132 d at 4°C (34). *Listeria monocytogenes* was detected after 1 yr in inoculated 1% dextrose broth containing 6% NaCl (37). At 4 or 10°C in BHI broth, the organism grew in the presence of 6% NaCl over 33 d; populations remained the same in 16% NaCl, and numbers declined slowly in 26% NaCl (16).

At 4 or 22°C, *L. monocytogenes* grew in skim milk and whey containing 6% NaCl (pH 6.20 and 5.65, respectively) but slowly decreased in skim milk and whey containing 12% NaCl (pH 6.00 and 5.50, respectively) (29). The same study found variation in salt tolerance between strains; strain Scott A was significantly more salt tolerant than strain California.

Substitution of KCl or a 55% NaCl/KCl for NaCl did not significantly alter survival of *L. monocytogenes* in rennet whey (20). In addition to its effect on survival, NaCl has been investigated for its effect on virulence.

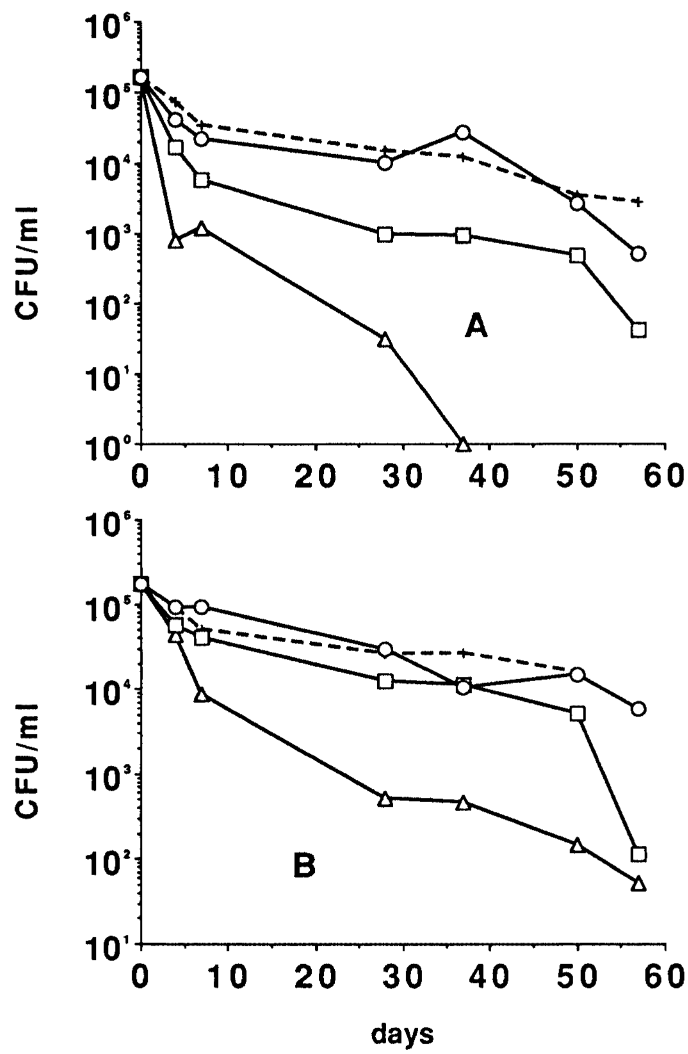

Figure 4. Survival of *Listeria monocytogenes* in commercial brines B3 (A) or M9 (B) with 0% (+), 0.1% (○), 0.5% (□), or 1% (△) added sodium benzoate at 4°C. The points are averages of duplicate samples.

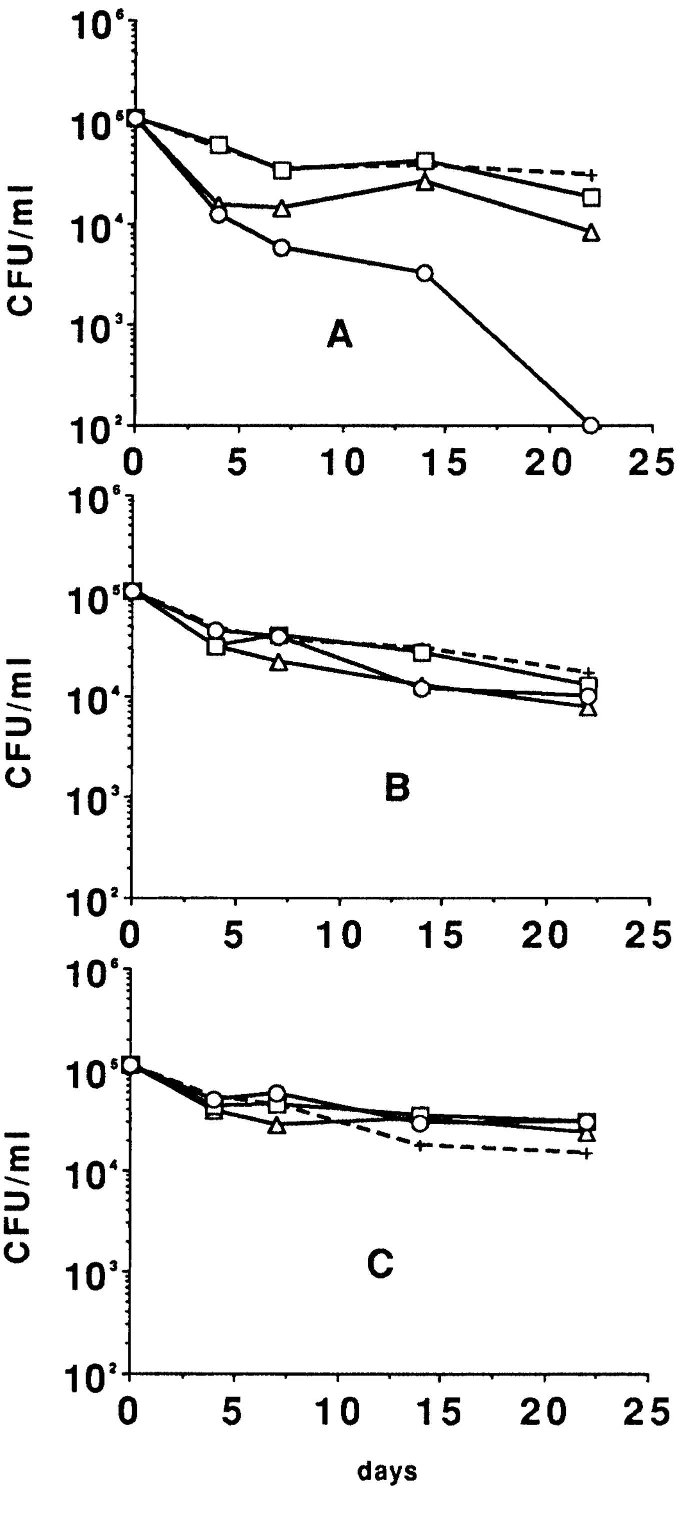

Figure 5. Survival of *Listeria monocytogenes* in commercial brines M1 (A), B2 (B), or S5 (C) after no extra filtration (+), filtration through coarse filter (△), filtration through coarse and 0.45-μm filters (□), or filtration through coarse, 0.45-μm, and 0.20-μm filters (○).

The virulence of *L. monocytogenes* was unaffected by growth in media containing 428 to 1112 m*M* (2.5 to 6.5%) NaCl (24) or 428 to 1112 m*M* (3.2 to 8.3%) KCl (25).

In the present study, *L. monocytogenes* was able to survive for over 200 d in many commercial brines with little decrease (≤1 log cfu/ml) in population in provolone and brick cheese brines at 4°C. There was no obvious correlation with survival of *L. monocytogenes* and brine pH, percentage of NaCl, microbial populations, mineral content, or nitrogen content. In general, inoculated *L. monocytogenes* was detected longer in commercial brines incubated at 4°C than at 12°C, which agrees with other studies that show greater survival in salt solutions at 4°C than at higher temperatures (14, 34).

Results of *L. monocytogenes* survival studies in commercial brines filtered in the laboratory before inoculation indicate that filtration of coarse particles has little effect on survival. However, filtration through 0.2-μm pore size filters (similar to UF in commercial brine systems) moderately decreased survival in one of three brines tested. This finding appears to differ from the *L. monocytogenes* survival in UF commercial brines in which the organism was detected at the last sampling time in 7 out of 13 brines incubated at 4°C.

The addition of sodium hypochlorite at unknown levels to five commercial brines at the plant level adversely affected survival of inoculated *L. monocytogenes*. Addition of sodium hypochlorite to two commercial brines at 10 to 100 ppm in the laboratory also strongly inhibited survival of *L. monocytogenes*. Sodium hypochlorite has been shown in previous research to be effective against *L. monocytogenes* in vitro and on stainless steel chips (23), confirming its effectiveness as a sanitizer on stainless steel surfaces. In other studies, available chlorine levels of 0.5 to 10 ppm (8), 1 ppm (9), >50 ppm (2), or 100 ppm (21) were effective against *L. monocytogenes*.

Sodium hypochlorite and other chlorinated sanitizers are widely used in the food and dairy industries. As described above, our results verify the antilisterial properties of sodium hypochlorite when added to cheese brines. However, addition of sodium hypochlorite to commercial cheese brines as an antimicrobial agent may face regulatory hurdles.

Other antimicrobials were antilisterial when added to two commercial cheese brines. Hydrogen peroxide at 0.005 to 0.02% strongly inhibited survival of the organism. In other research, *L. monocytogenes* was completely inactivated at 9 h, when added to sterilized milk containing 0.0495% hydrogen peroxide (6). However, hydrogen peroxide is not a permitted additive in milk and most dairy products in most developed countries.

At the relatively high level of 1%, potassium sorbate and sodium benzoate each slightly inhibited *L. monocy-*

togenes survival in two commercial brines. Sodium benzoate at levels of 0.15 to 0.3% has been shown to inactivate *L. monocytogenes* in tryptose broth with more effective inhibition at pH 5.0 than 5.6 (10). Normal usage level of sodium benzoate in foods is up to 0.2 to 0.3% (10). Thus, potassium sorbate or sodium benzoate could be used as antilisterial agents in commercial brines, but the cost effectiveness of adding them at high levels (1%) is questionable, and their long-term stability in the high-salt and low-pH environment of brines is not well defined.

Listeria monocytogenes has been shown to be capable of extended survival in some commercial cheese brines. Considering the prevalence of *L. monocytogenes* in cheese plant environments, the ability of the organism to leach from contaminated cheese into the brine, the practical limits of significantly lowering brine pH, and limited feasability of adding antimicrobials to the brine, it would seem that there is a limited, but real, possiblility of commercial cheese brines becoming a reservoir for *L. monocytogenes*. The use of good management practices, especially proper sanitation of the cheese plant environment, would seem to be a key in preventing brine contamination by the organism.

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