FRI BRIEFINGS

LITERATURE SURVEY OF THE VARIOUS TECHNIQUES USED IN *LISTERIA* INTERVENTION

Recalls, illnesses, and deaths associated with *Listeria* in food products have been reported over the last year. These incidences have increased the awareness that additional techniques may be needed for controlling *Listeria* in food processing plants and, especially, those producing Ready to Eat (RTE) products. Additionally, the Food Safety and Inspection Service (FSIS) of the USDA has issued performance standards that amend Federal meat and poultry inspection regulations of certain RTE meat and poultry products, effective March 8, 1999 [*Federal Register* 64(3):732–749, January 6, 1999]. The new performance standards spell out the objective level of food safety performance that establishments must meet. However, they allow establishments to develop and implement processing procedures customized to the nature and volume of their production. This makes this review of *Listeria* intervention techniques very timely.

The American Meat Institute (AMI) Foundation encouraged and financially contributed to the preparation of this brief.

page

USE OF ORGANIC ACIDS TO CONTROL *LISTERIA* IN MEAT

A low pH (acidic) environment has an adverse effect on the growth of *Listeria monocytogenes* but it is not only the specific pH of the medium which is important but also the type of acid, temperature, and other antimicrobial compounds which are present (*7*). Several researchers have noted that, in culture media, acetic acid has more potent antilisterial effects than lactic acid, which, in turn, is more inhibitory than hydrochloric acid (*1,19,20,36*). Although similar concentrations of citric and lactic acids reduce the pH of tryptic soy broth more than acetic acid does, addition of acetic acid results in greater cell destruction (*19*). Malic acid, the predominant organic acid in apples, is not as effective as lactic acid in suppressing growth of *L. monocytogenes* (*4*). Sodium diacetate (a mixture of acetic acid and sodium acetate) also significantly inhibits the growth of *L. monocytogenes* in broth cultures (*32*). Several experiments in culture media demonstrated that inhibitory effects of an acid are greater at lower temperatures (*5,6,13,16,17,31*).

Other factors, such as the presence of salt and other compounds used as preservatives, may modify the effects of organic acids on *L. monocytogenes* (*6,16,21, 31*), and several models have been developed to describe these interactions (*5,17,26*). These models may provide useful estimates of the relative importance of different factors and the magnitude of inhibition to be expected but they may overestimate or underestimate the effects on *L. monocytogenes* in meat, such as bologna (*17*) and sausage (*26*).

Organic acids can interact with other preservatives to enhance their effects. Acetic and lactic acids enhance the antilisterial effects of monolaurin (*25,27,28*). Lactic acid increased the susceptibility of *L. monocytogenes* to heat shock in culture media (*20*). But no effect on thermal tolerance was observed in ground pork (*39*).

However, it should be noted that the effects of organic acids are not always positive in terms of food safety. Listeriae which are exposed to these acids and survive may repair themselves during storage at low temperatures and begin to multiply if other barriers are not present (*9,14,29*). Exposure to acid also induces stress responses in listeriae which make the bacteria more tolerant of more acidity, ethanol, and hydrogen peroxide (*22*).

Antilisterial effects of organic acids have been examined in several types of meats: raw, cooked, and cured. Since carcass meat may be contaminated with *L. monocytogenes* during slaughter and packaging into retail cuts of meat, solutions of organic acids have been tested as washes or dips for removing listeriae from meat and/or inhibiting its growth during refrigerated storage. When lactic or acetic acids (1.5–4%) were sprayed on contaminated beef carcass or beef trim, large numbers of inoculated *L. monocytogenes* persisted and grew on the meat stored under refrigeration (*10,11*). On the other hand, if the beef was sprayed with 2% lactic or acetic acid before it was contaminated with *L. monocytogenes*, the residual activity of the acids suppressed the growth of the bacteria (*12*). Organic acids (1–3%) used as dips are usually more efficacious than carcass washes because some residual activity remains on the meat. These acid concentrations generally cause no adverse effect on the sensory properties of the meat. *Listeria monocytogenes* and *E. coli*, however, are more resistant to acid treatments than *Yersinia* and *Salmonella* (*14,34*). Both lactic acid (1.7%) and acetic acid (2%) reduced *L. monocytogenes* populations on lean beef tissue by 2–3 logs for up to 7 days (*33*). In other experiments with raw beef, 2% fumaric acid was found to be a more effective antilisterial agent than 1% acetic or lactic acid (*30*). When lean pork tissue and pork fat were artificially inoculated with *L. monocytogenes* and then dipped in 3% lactic acid or water for 15 sec, numbers of listeriae were reduced by 1–2 logs for the lean meat and up to 7 logs for the fat during 15 days of refrigerated storage (*14*). The more potent effects observed for pork fat were probably due to the fact that acid-treated fat was approximately 2.5 pH units lower than acid-treated lean tissue. A similar effect was observed in pork liver sausage with 22–67% fat treated with propionate or lactate: at higher fat levels, the kill was approximately 2–3 times greater (*18*).

The best treatment for artificially contaminated raw chicken legs was reported to be a wash with a 10% lactic acid/sodium lactate buffer, pH 3.0 followed by packaging in 90% carbon dioxide, 10% oxygen. This procedure extended the shelf life of the chicken from 6 days to 17 days. Chicken treated with the lactate buffer without modified atmosphere packaging had a shelf life of 10 days (*40*).

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison
November 1999

Control of *Listeria* in Meat by Organic Acids

Artificial contamination of frankfurters with *L. monocytogenes* followed by a 2 min dip in 1% lactic, acetic, tartaric, or citric acids resulted in a 1–2 log kill of the bacteria. However, surviving bacteria started growing during refrigerated storage. A dip in 5% acetic or lactic acids not only killed *L. monocytogenes* but prevented its regrowth during 90 days storage (*29*).

Addition of 1.8% or 2% lactic acid to raw or cooked ground beef did not appreciably affect the survival and growth of *L. monocytogenes* (*15*,*37*). Data from another experiment indicated that lactic acid slightly reduced the thermal tolerance of *L. monocytogenes* in ground beef (*23*). Sodium diacetate (0.3%) delayed growth of *L. monocytogenes* in turkey slurry (*31*).

Sodium lactate (4%) was reported to suppress the growth of *L. monocytogenes* in cooked strained beef (*8*) and beef roasts (*24*). In both cases, however, there were viable listeriae left in the meat during refrigeration. *L. monocytogenes* inoculated onto cooked chicken which was treated with lactate were observed to have a longer lag phase but were still able to grow during storage (*2*). Brines containing monolaurin and lactate pumped into beef roasts (microwave-ready beef roasts) enabled a greater kill of *L. monocytogenes* during cooking in bags in water baths than brines with only lactate (*35*).

Cured meats, such as sausage, ham, and frankfurters, which contain salt and other preservatives, are more susceptible to the listericidal effects of organic acids (*3*,*17*,*18*,*26*, *29*,*38*)

REFERENCES

1. Ahamad N, Marth EH. Acid-injury of *Listeria monocytogenes*. J. Food Protect. 1990; 53(1):26–29.
2. Barakat RK, Harris LJ. Growth of *Listeria monocytogenes* and *Yersinia enterocolitica* on cooked modified-atmosphere-packaged poultry in the presence and absence of a naturally occurring microbiota. Appl. Environ. Microbiol. 1999; 65(1):342–345
3. Blom H, Nerbrink E, Dainty R, Hagtvedt T, Borch E, Nissen H, Nesbakken T. Addition of 2.5% lactate and 0.25% acetate controls growth of *Listeria monocytogenes* in vacuum-packed, sensory-acceptable servelat sausage and cooked ham stored at 4ºC. Int. J. Food Microbiol. 1997; 38(1):71–76.
4. Buchanan RL, Golden MH. Interactions between pH and malic acid concentration on the inactivation of *Listeria monocytogenes*. J. Food Safety 1998; 18(1):37–48.
5. Buchanan RL, Golden MH, Phillips J G. Expanded models for the non-thermal inactivation of *Listeria monocytogenes*. J. Appl. Microbiol. 1997; 82(5):567–577.
6. Buchanan RL, Stahl HG, Whiting RC. Effects and interactions of temperature, pH, atmosphere, sodium chloride, and sodium nitrite on the growth of *Listeria monocytogenes*. J. Food Protect. 1989; 52(12):844–851.
7. Buchanan RL, Golden MH, Whiting RC. Differentiation of the effects of pH and lactic or acetic acid concentration on the kinetics of *Listeria monocytogenes* inactivation. J. Food Protect. 1993; 56(6):474–478.
8. Chen N and Shelef LA. Relationship between water activity, salts of lactic acid, and growth of *Listeria monocytogenes* in a meat model system. J. Food Protect. 1992; 55(8):574–578.
9. Cheroutre-Vialette M, Lebert I, Hebraud M, Labadie JC, Lebert A. Effects of pH or a_w stress on growth of *Listeria monocytogenes*. Int. J. Food Microbiol. 1998; 42(1–2):71–77.
10. Conner DE, Kotrola JS, Mikel WB, Tamblyn KC. Effects of acetic-lactic acid treatments applied to beef trim on populations of *Escherichia coli* O157-H7 and *Listeria monocytogenes* in ground beef. J. Food Protect. 1997; 60(12):1560–1563.
11. Dorsa WJ, Cutter CN, Siragusa GR. Effects of acetic acid, lactic acid and trisodium phosphate on the microflora of refrigerated beef carcass surface tissue inoculated with *Escherichia coli* O157-H7, *Listeria innocua*, and *Clostridium sporogenes*. J. Food Protect. 1997; 60(6):619–624.
12. Dorsa WJ, Cutter CN, Siragusa GR. Long-term bacterial profile of refrigerated ground beef made from carcass tissue, experimentally contaminated with pathogens and spoilage bacteria after hot water, alkaline, or organic acid washes. J. Food Protect. 1998; 61(12):1615–1622.
13. Gill CO, Greer GG, Dilts BD. The aerobic growth of *Aeromonas hydrophila* and *Listeria monocytogenes* in broths and on pork. Int. J. Food Microbiol. 1997; 35(1):67–74.
14. Greer GG, Dilts BD. Lactic acid inhibition of the growth of spoilage bacteria and cold tolerant pathogens on pork. Int. J. Food Microbiol. 1995; 25(2):141–151.
15. Harmayani E, Sofos JN, Schmidt GR. Fate of *Listeria monocytogenes* in raw and cooked ground beef with meat processing additives. Int. J. Food Microbiol. 1993; 18(3):223–232.
16. Houtsma PC, Dewit JC, Rombouts FM. Minimum inhibitory concentration (MIC) of sodium lactate and sodium chloride for spoilage organisms and pathogens at different pH values and temperatures. J. Food Protect. 1996; 59(12):1300–1304.
17. Houtsma PC, Kant-Muermans ML, Rombouts FM, Zwietering MH. Model for the combined effects of temperature, pH, and sodium lactate on growth rates of *Listeria innocua* in broth and bologna-type sausages. Appl. Environ. Microbiol. 1996; 62(5):1616–1622.
18. Hu AC, Shelef LA. Influence of fat content and preservatives on the behavior of *Listeria monocytogenes* in beaker sausage. J. Food Safety 1996; 16(3):175–181.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

Control of *Listeria* in Meat by Organic Acids

19. Ita PS, Hutkins RW. Intracellular pH and survival of *Listeria monocytogenes* Scott A in tryptic soy broth containing acetic, lactic, citric, and hydrochloric acids. J. Food Protect. 1991; 54(1):15–19.

20. Jorgensen F, Hansen TB, Knochel S. Heat shock-induced thermotolerance in *Listeria monocytogenes* 13-249 is dependent on growth phase, pH and lactic acid. Food Microbiol. 1999; 16(2):185–194.

21. Kamat AS, Nair, PM. Identification of *Listeria innocua* as a biological indicator for inactivation of *L. monocytogenes* by some meat processing treatments. Food Sci. Technol. Lebensm. Wiss. Technol. 1996; 29(8):714–720.

22. Lou YQ, Yousef AE. Adaptation to sublethal environmental stresses protects *Listeria monocytogenes* against lethal preservation factors. Appl. Environ. Microbiol. 1997; 63(4):1252–1255.

23. McMahon CMM, Doherty AM, Sheridan JJ, Blair IS, McDowell DA, Hegarty T. Synergistic effect of heat and sodium lactate on the thermal resistance of *Yersinia enterocolitica* and *Listeria monocytogenes* in minced beef. Lett. Appl. Microbiol. 1999; 28(5):340–344.

24. Miller RK and Acuff GR. Sodium lactate affects pathogens in cooked beef. J. Food Sci. 1994; 59(1):15–19.

25. Monk JD, Beuchat LR, Hathcox AK. Inhibitory effects of sucrose monolaurate, alone and in combination with organic acids, on *Listeria monocytogenes* and *Staphylococcus aureus*. J. Appl. Bacteriol. 1996; 81(1):7–18.

26. Nerbrink E, Borch E, Blom H, Nesbakken T. A model based on absorbance data on the growth rate of *Listeria monocytogenes* and including the effects of pH, NaCl, Na–lactate and Na-acetate. Int. J. Food Microbiol. 1999; 47(1–2):99–109.

27. Oh DH, Marshall DL. Monolaurin and acetic acid inactivation of *Listeria monocytogenes* attached to stainless steel. J. Food Protect. 1996; 59(3):249–252.

28. Oh DH, Marshall DL. Effect of pH on the minimum inhibitory concentration of monolaurin against *Listeria monocytogenes*. J. Food Protect. 1992; 55(6):449–450.

29. Palumbo SA, Williams AC. Control of *Listeria monocytogenes* on the surface of frankfurters by acid treatments. Food Microbiol. 1994; 11(4):293–300.

30. Podolak RK, Zayas JF, Kastner CL, Fung DYC. Inhibition of *Listeria monocytogenes* and *Escherichia coli* O157-H7 on beef by application of organic acids. J. Food Protect. 1996; 59(4):370–373.

31. Schlyter JH, Glass KA, Loeffelholz J, Degnan AJ, Luchansky, JB. The effects of diacetate with nitrite, lactate, or pediocin on the viability of *Listeria monocytogenes* in turkey slurries. Int. J. Food Microbiol. 1993; 19(4):271–281.

32. Shelef LA, Addala L. Inhibition of *Listeria monocytogenes* and other bacteria by sodium diacetate. J. Food Safety 1994; 14(2):103–115.

33. Siragusa GR, Dickson JS. Inhibition of *Listeria monocytogenes, Salmonella typhimurium* and *Escherichia coli* 0157:H7 on beef muscle tissue by lactic or acetic acid contained in calcium alginate gels. J. Food Safety 1993; 13(2):147–158.

34. Smulders FJM, Greer GG. Integrating microbial decontamination with organic acids in HACCP programmes for muscle foods — prospects and controversies. Int. J. Food Microbiol. 1998; 44(3):149–169.

35. Unda JR, Molins RA, Walker HW. *Clostridium sporogenes* and *Listeria monocytogenes*: Survival and inhibition in microwave-ready beef roasts containing selected antimicrobials. J. Food Sci. 1991; 56(1):198–205.

36. Vasseur C, Baverel L, Hebraud M, Labadie J. Effect of osmotic, alkaline, acid or thermal stresses on the growth and inhibition of *Listeria monocytogenes*. J. Appl. Microbiol. 1999; 86(3):469–476.

37. Vignolo G, Fadda S, de Kairuz MN, Holgado APD, Oliver G. Effects of curing additives on the control of *Listeria monocytogenes* by lactocin 705 in meat slurry. Food Microbiol. 1998; 15(3):259–264.

38. Weaver RA, Shelef LA. Antilisterial activity of sodium, potassium or calcium lactate in pork liver sausage. J. Food Safety 1993; 13(2):133–146.

39. Yen LC, Sofos JN, Schmidt GR. Destruction of *Listeria monocytogenes* by heat in ground pork formulated with kappa-carrageenan, sodium lactate and the algin/calcium meat binder. Food Microbiol. 1992; 9(3):223–230.

40. Zeitoun AAM, Debevere JM. Inhibition, survival and growth of *Listeria monocytogenes* on poultry as influenced by buffered lactic acid treatment and modified atmosphere packaging. Int. J. Food Microbiol. 1991; 14:161–170.

USE OF OTHER PRESERVATIVES TO CONTROL *LISTERIA* IN MEAT

Since *Listeria monocytogenes* can grow on a variety of processed meat products at refrigeration temperatures (*9*), a variety of chemicals which destroy or limit the growth of harmful microbes have been tested for the preservation of meat. Many of these compounds are well known and their effects on various bacteria and on meat quality have been thoroughly investigated; others have been introduced recently and are not as well studied. Some compounds are not very potent by themselves but in combination with other preservatives or storage conditions can suppress the growth of foodborne pathogens. Several researchers have developed models which describe the effects of different combinations of preservatives on the growth of *L. monocytogenes* in laboratory media (*2*,*8*,*26*,*31*). Although these models are useful, growth of *L. monocytogenes* in meat nearly always differs from that in culture media.

Sodium chloride (NaCl). NaCl in growth media or foods can be a source of osmotic stress by decreasing water activity (a_w). However, *L. monocytogenes* is remarkably salt-tolerant and able to withstand higher salt concentrations than *Salmonella* spp. and *Yersinia* spp. (*13*). In an experiment to determine the antilisterial effects of brine solutions which could be used as dips, *L. monocytogenes* easily survived 6 hours at 10°C in solutions containing 6, 16, or 26% sodium chloride (*15*). *Listeria monocytogenes* even grew in the 6% brine solution (*15*) and in meat peptone media containing 8% NaCl (*40*). The presence of sodium chloride in growth media also partially protects *L. monocytogenes* from other stresses such as heat in ground pork (*45*), lactocin 705 in minced beef slurry (*41*), and hydrogen peroxide in culture media (*21*).

Although *L. monocytogenes* is halotolerant, salt is a stress and does depress growth rates (*4*,*40*). In combination with other compounds used in curing meats, NaCl is one factor contributing to the destruction or inhibition of *L. monocytogenes* (*3*,*8*,*17*,*26*,*31*).

Nitrite. Nitrite alone is also not a very effective antilisterial agent. In turkey slurries (pH 6.2), 30 ppm sodium nitrite was unable to inhibit the growth of *L. monocytogenes* at 4 or 25°C (*35*). In beef slurries, 800 ppm was required to inhibit growth of *L. monocytogenes* (*41*). However, as with salt, in the presence of other curing agents (*8*,*26*,*31*,*44*) or lactocin 705 (*41*), nitrite can contribute to the suppression of *L. monocytogenes* at refrigeration temperatures.

Trisodium phosphate (TSP). Trisodium phosphate has been used for decontamination of poultry carcasses (*34*) and can reduce bacterial contaminants by 1–2 logs. Spraying of TSP on beef carcass tissue contaminated with *L. monocytogenes* removed 1.3 log of cells but by the 7th day of cold storage, the remaining bacteria started to grow (*6*). Use of 10% TSP as a 15 sec dip removed only about 39% and 81% of *L. monocytogenes* at 10°C and 4°C, respectively (*5*). In other experiments, in which *L. monocytogenes* was suspended on solutions of TSP, exposure to 8% TSP for at least 10 min was required to reduce bacterial numbers by at least 1 log (*36*). *Escherichia coli* O157:H7, *Campylobacter jejuni* and *Salmonella typhimurium* were all more sensitive than *L. monocytogenes* to TSP.

Smoke/Liquid Smoke. Smoking of meat and fish is a well-known preservation technique and has been shown to inhibit the growth of *L. monocytogenes* (*27*,*32*). Several experiments have also documented the antilisterial effects of liquid smoke additives. Of 5 Red Arrow smoke products evaluated, CharSol-10 was the most effective against *L. monocytogenes* and reduced viable cells on the surface of beef franks by >99.9% after 72 hours storage at 4°C (*23*). Another product, CharSol Supreme, also had potent antilisterial effects in wiener exudate (*7*). Analysis of this product revealed that its active ingredient was isoeugenol and that this compound was more effective in the presence of acetic acid at pH 5.8. Experiments with 7 commercial smoke preparations used in Spain indicated that some were better antilisterial agents than others and that the most potent had higher concentrations of phenols (*37*).

Plant Extracts. A variety of herbs and spices have been tested for their efficacy in suppressing the growth of *L. monocytogenes* in culture media. Plant extracts exhibiting antilisterial activity include: hop extracts (*20*), eugenol (*1*,*10*,*11*), pimento leaf (*10*,*11*), horseradish distillates (*43*), rosemary (*21*,*30*), cloves (*21*,*30*), cin-

Control of *Listeria* in Meat by Other Preservatives

namic acid (*19,33*), furanocoumarins (*38*), and carvacol (*18*). Numerous other plant extracts have been tested but results were not always consistent. (*10,18,21*). Different commercial samples of plant essential oils and different varieties of the same herbs may exhibit differences in antilisterial potency because of varying amounts of critical compounds. Some plant extracts were also found to be effective against *Listeria* spp. in meat including rosemary in ready-to-eat pork liver sausage (*30*), horseradish distillates on roast beef (*43*), and eugenol and pimento leaf on refrigerated cooked beef (*11*). It should be noted that *L. monocytogenes* was usually less sensitive to these extracts in meat (compared to culture media) and sensitivity also varied with fat content of the meat. For hop extracts tested in dairy products, antimicrobial activity was higher in lower fat meats (*20*).

Monolaurin and other monoglycerides. Several monoglycerides (glycerol with one esterified fatty acid) are effective inhibitors of *L. monocytogenes* in culture media (*25,28,29,42*) and in foods. In beef frank slurries (pH 5.0 and 5.5), monocaprin, monolaurin and coconut monoglycerides individually all inhibited the growth of *L. monocytogenes* (*42*). These individual compounds were not as effective in turkey frank slurries but combinations of monoglycerides were effective. Brines containing monolaurin and lactate pumped into beef roasts (microwave-ready beef roasts) enabled a greater kill of *L. monocytogenes* during cooking in bags in water baths than brines without monolaurin (*39*). Monolaurin appeared to be a more potent antimicrobial at lower temperatures and pH values (*25,29,42*). Also, planktonic cells of *L. monocytogenes* were more susceptible to monolaurin than cells attached to stainless steel surfaces (*28*).

Chelators (Citrate and EDTA). Chelators, which bind metal ions, are not by themselves lethal to *L. monocytogenes* in the concentrations used in foods (*46*). However, these compounds interact with other preservatives and sometimes aid in suppressing the growth of *L. monocytogenes* in meats (*1,25,31*). In other cases, for example EDTA combined with nisin, the opposite occurs and EDTA reduces the antimicrobial effects of nisin (*46*).

Lysozyme. Hen egg white lysozyme suppressed the growth of *L. monocytogenes* in fresh pork sausage (bratwurst) for 2–3 weeks (*16*).

Sorbate (Sorbic acid). Experiments using culture media revealed that *L. monocytogenes* was more susceptible to sorbate at lower pH (pH 5 vs pH 6) and at lower temperatures (5°C vs 30°C) (*24*). In beaker sausage, sorbate was also a more effective inhibitor of *L. monocytogenes* at lower temperatures (*14*). Fat content of the sausage did not affect the potency of sorbate at 4°C, but at 10°C sorbate was a more effective in sausages containing 67% fat as compared to 22% fat.

Other additives. Minimal inhibitory concentrations of **methyl paraben (p-hydroxybenzoate)** for growth of *L. monocytogenes* in culture media were lower at pH 5 than at pH 6 and at 5°C than at 30°C. Under similar conditions, methyl paraben was a more potent inhibitor of *L. monocytogenes* than sorbate (*24*). **Sodium erythorbate** did not appear to be an effective antilisterial agent in raw or cooked ground beef (*12*).

REFERENCES

1. Blaszyk M, Holley RA. Interaction of monolaurin, eugenol and sodium citrate on growth of common meat spoilage and pathogenic organisms. Int. J. Food Microbiol. 1998; 39(3):175–183.

2. Buchanan RL, Golden MH, Phillips JG. Expanded models for the non-thermal inactivation of *Listeria monocytogenes*. J. Appl. Microbiol. 1997; 82(5):567–577.

3. Buchanan RL, Stahl HG, Whiting RC. Effects and interactions of temperature, pH, atmosphere, sodium chloride, and sodium nitrite on the growth of *Listeria monocytogenes*. J. Food Protect. 1989; 52(12):844–851.

4. Cheroutre-Vialette M, Lebert I, Hebraud M, Labadie JC, Lebert A. Effects of pH or a_w stress on growth of *Listeria monocytogenes*. Int. J. Food Microbiol.1998; 42(1–2):71–77.

5. Deledesma AMR, Riemann HP, Farver TB. Short-time treatment with alkali and/or hot water to remove common pathogenic and spoilage bacteria from chicken wing skin. J. Food Protect. 1996; 59(7):746–750.

6. Dorsa WJ, Cutter CN, Siragusa GR. Effects of acetic acid, lactic acid and trisodium phosphate on the microflora of refrigerated beef carcass surface tissue inoculated with *Escherichia coli* O157-H7, *Listeria innocua*, and *Clostridium sporogenes*. J. Food Protect. 1997; 60(6):619–624.

7. Faith NG, Yousef AE, Luchansky JB. Inhibition of *Listeria monocytogenes* by liquid smoke and isoeugenol, a phenolic component found in smoke. J. Food Safety 1992; 12(4):303–314.

8. Fernandez PS, George SM, Sills CC, Peck MW. Predictive model of the effect of CO_2, pH, temperature and NaCl on the growth of *Listeria monocytogenes*. Int. J. Food Microbiol. 1997; 37(1):37–45.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

Control of *Listeria* in Meat by Other Preservatives

9. Glass KA, Doyle MP. Fate of *Listeria monocytogenes* in processed meat products during refrigerated storage. Appl. Environ. Microbiol. 1989; 55(6)1565–1569.

10. Hao YY, Brackett RE, Doyle MP. Efficacy of plant extracts in inhibiting *Aeromonas hydrophila* and *Listeria monocytogenes* in refrigerated, cooked poultry. Food Microbiol. 1998; 15(4):367–378.

11. Hao YY, Brackett RE, Doyle MP. Inhibition of *Listeria monocytogenes* and *Aeromonas hydrophila* by plant extracts in refrigerated cooked beef. J. Food Protect. 1998; 61(3):307–312.

12. Harmayani E, Sofos JN, Schmidt GR. Fate of *Listeria monocytogenes* in raw and cooked ground beef with meat processing additives. Int. J. Food Microbiol. 1993; 18(3):223–232.

13. Houtsma PC, Dewit JC, Rombouts FM. Minimum inhibitory concentration (MIC) of sodium lactate and sodium chloride for spoilage organisms and pathogens at different pH values and temperatures. J. Food Protect. 1996; 59(12):1300–1304.

14. Hu AC, Shelef LA. Influence of fat content and preservatives on the behavior of *Listeria monocytogenes* in beaker sausage. J. Food Safety. 1996; 16(3):175–181.

15. Hudson JA. Efficacy of high sodium chloride concentrations for the destruction of *Listeria monocytogenes*. Lett. Appl. Microbiol. 1992; 14(4):178–180.

16. Hughey VL, Wilger PA, Johnson EA. Antibacterial activity of hen egg white lysozyme against *Listeria monocytogenes* Scott A in foods. Appl. Environ. Microbiol. 1989; 55(3):631–638.

17. Kamat AS, Nair PM. Identification of *Listeria innocua* as a biological indicator for inactivation of *L. monocytogenes* by some meat processing treatments. Food Sci. Technol. Lebensm. Wiss. Technol. 1996; 29(8):714–720.

18. Kim JM, Marshall MR, Wei C. Antibacterial activity of some essential oil components against five foodborne pathogens. J. Agric. Food Chem. 1995; 43(11):2839–2845.

19. Kouassi Y, Shelef LA. Inhibition of *Listeria monocytogenes* by cinnamic acid — possible interaction of the acid with cysteinyl residues. J. Food Safety 1998; 18(3):231–242.

20. Larson AE, Yu RRY, Lee OA, Price S, Haas GJ, Johnson EA. Antimicrobial activity of hop extracts against *Listeria monocytogenes* in media and in food. Int. J. Food Microbiol. 1996; 33(2–3):195–207.

21. Lis-Balchin M, Deans SG. Bioactivity of selected plant essential oils against *Listeria monocytogenes*. J. Appl. Microbiol. 1997; 82(6):759–762.

22. Lou YQ, Yousef AE. Adaptation to sublethal environmental stresses protects *Listeria monocytogenes* against lethal preservation factors. Appl. Environ. Microbiol. 1997; 63(4):1252–1255.

23. Messina MC, Ahmad HA, Marchello JA, Gerba CP, Paquette MW. The effect of liquid smoke on *Listeria monocytogenes*. J. Food Protect. 1988; 51(8):629–631, 638.

24. Moir CJ, Eyles MJ. Inhibition, injury, and inactivation of four psychrotrophic foodborne bacteria by the preservatives methyl p-hydroxybenzoate and potassium sorbate. J. Food Protect. 1992; 55(5):360–366.

25. Monk JD, Beuchat LR, Hathcox AK. Inhibitory effects of sucrose monolaurate, alone and in combination with organic acids, on *Listeria monocytogenes* and *Staphylococcus aureus*. J. Appl. Bacteriol. 1996; 81(1):7–18.

26. Nerbrink E, Borch E, Blom H, Nesbakken T. A model based on absorbance data on the growth rate of *Listeria monocytogenes* and including the effects of pH, NaCl, Na-lactate and Na-acetate. Int. J. Food Microbiol. 1999; 47(1–2):99–109.

27. Niedziela JC, MacRae M, Ogden ID, Nesvadba P. Control of *Listeria monocytogenes* in salmon — antimicrobial effect of salting, smoking and specific smoke compounds. Food Sci. Technol.-Lebensm. Wiss. Technol. 1998; 31(2):155–161.

28. Oh DH, Marshall DL. Monolaurin and acetic acid inactivation of *Listeria monocytogenes* attached to stainless steel. J. Food Protect. 1996; 59(3):249–252.

29. Oh DH, Marshall DL. Effect of pH on the minimum inhibitory concentration of monolaurin against *Listeria monocytogenes*. J. Food Protect. 1992; 55(6):449–450.

30. Pandit VA, Shelef LA. Sensitivity of *Listeria monocytogenes* to rosemary (*Rosmarinus officinalis* L.). Food Microbiol. 1994; 11(1):57–63.

31. Parente E, Giglio, MA, Ricciardi A, Clementi F. The combined effect of nisin, leucocin F10, pH, NaCl and EDTA on the survival of *Listeria monocytogenes* in broth. Int. J. Food Microbiol. 1998; 40(1–2):65–75.

32. Poysky FT, Paranjpye RN, Peterson ME, Pelroy GA, Guttman AE, Eklund MW. Inactivation of *Listeria monocytogenes* on hot-smoked salmon by the interaction of heat and smoke or liquid smoke. J. Food Protect. 1997; 60(6):649–654.

33. Ramos-Nino ME, Clifford MN, Adams MR. Quantitative structure activity relationship for the effect of benzoic acids, cinnamic acids and benzaldehydes on *Listeria monocytogenes*. J. Appl. Bacteriol. 1996; 80(3):303–310.

34. Salvat G, Coppen P, Allo JC, Fenner S, Laisney MJ, Toquin, MT, Humbert F, ColinP. Effects of Avgard(tm) treatment on the microbiological flora of poultry carcases. Brit. Poultry Sci. 1997; 38(5):489–498.

35. Schlyter JH, Glass KA, Loeffelholz J, Degnan AJ, Luchansky JB. The effects of diacetate with nitrite, lactate, or pediocin on the viability of *Listeria monocytogenes* in turkey slurries. Int. J. Food Microbiol. 1993; 19(4):271–281

36. Somers EB, Schoeni JL, Wong ACL. Effect of trisodium phosphate on biofilm and planktonic cells of *Campylobacter jejuni*, *Escherichia coli* O157:H7, *Listeria monocytogenes* and *Salmonella typhimurium*. Int. J. Food Microbiol. 1994; 22:269–276.

37. Suñen E. Minimum inhibitory concentration of smoke wood extracts against spoilage and pathogenic micro-organ-

isms associated with foods. Lett. Appl. Microbiol. 1998; 27(1):45–48.

38. Ulate-Rodriguez J, Schafer HW, Zottola EA, Davidson PM. Inhibition of *Listeria monocytogenes, Escherichia coli* O157-H7, and *Micrococcus luteus* by linear furanocoumarins in culture media. J. Food Protect. 1997; 60(9):1046–1049.

39. Unda JR, Molins RA, Walker HW. *Clostridium sporogenes* and *Listeria monocytogenes*: Survival and inhibition in microwave-ready beef roasts containing selected antimicrobials. J. Food Sci. 1991; 56(1):198–205.

40. Vasseur C, Baverel L, Hebraud M, Labadie J. Effect of osmotic, alkaline, acid or thermal stresses on the growth and inhibition of *Listeria monocytogenes*. J. Appl. Microbiol. 1999; 86(3):469–476.

41. Vignolo G, Fadda S, de Kairuz MN, Holgado APD, Oliver G. Effects of curing additives on the control of *Listeria monocytogenes* by lactocin 705 in meat slurry. Food Microbiol. 1998; 15(3):259–264.

42. Wang LL, Johnson EA. Control of *Listeria monocytogenes* by monoglycerides in foods. J. Food Protect. 1997; 60(2):131–138.

43. Ward SM, Delaquis PJ, Holley RA, Mazza G. Inhibition of spoilage and pathogenic bacteria on agar and precooked roast beef by volatile horseradish distillates. Food Res. Int. 1998; 31(1):19–26.

44. Yen LC, Sofos JN, Schmidt GR. Destruction of *Listeria monocytogene*s by heat in ground pork formulated with kappa-carrageenan, sodium lactate and the algin/calcium meat binder. Food Microbiol. 1992; 9 (3):223–230.

45. Yen LC, Sofos JN, Schmidt GR. Effect of meat curing ingredients on thermal destruction of *Listeria monocytogenes* in ground pork. J. Food Protect. 1991; 54(6):408–412.

46. Zhang SS, Mustapha A. Reduction of *Listeria monocytogenes* and *Escherichia coli* O157:H7 numbers on vacuum-packaged fresh beef treated with nisin or nisin combined with EDTA. J. Food Protect. 1999; 62(10):1123–1127.

USE OF BACTERIOCINS TO CONTROL *LISTERIA* IN MEAT

Bacteriocins are proteinaceous, antimicrobial compounds produced by many kinds of bacteria. Attempts to harness these compounds to control *Listeria monocytogenes* in meats have taken two approaches: (a) Add the bacteriocin directly to the food in a purified or partially purified form. (b) Add the bacteriocin-producing bacteria to the meat so they will grow and produce bacteriocins in situ. Some recent reviews summarize results of experiments using bacteriocins to control *L. monocytogenes* in foods and discussed modes of action of these compounds, factors affecting their effectiveness, and development of resistance in *L. monocytogenes* (*2,15,26*). In particular, Muriana (*26*) discusses the use of bacteriocins for controlling *L. monocytogenes* and includes some earlier references which are not cited in this report.

Bacterial Cultures. Since lactobacilli are known to produce many different bacteriocins and some are also used in starter cultures for sausage production, addition of these bacteriocin producers has been effective in reducing *L. monocytogenes* populations in many fermented meats (*8,12,14,30,37*). Some bacteriocinogenic strains do not grow well at refrigeration temperatures and thus may be more useful in controlling listeriae at temperature abuse conditions rather than in refrigerated storage (*4*). Other bacteria produce higher levels of bacteriocins at low temperatures (*5*). Bacteriocinogenic strains have also been used to control spoilage organisms (*20*).

Lactobacilli also produce lactic acid which acidifies the meat and, in some cases, antilisterial effects of lactobacilli have been traced to lactic acid rather than to bacteriocins (*18*).

Lactocin 705. Lactocin 705, produced by *Lactobacillus casei* CRL 705, exerted a moderate inhibitory effect on the growth of *L. monocytogenes* in minced beef slurry (*36*). Further experiments with sodium chloride, nitrite, and lactate added to minced beef demonstrated that these curing salts reduced the effectiveness of lactocin 705 (*35*).

Nisin. Nisin is currently being used for the preservation of some foods because of its GRAS status and well-known antilisterial effects. Several factors affecting the inhibitory activity of nisin were investigated in broth cultures (*28*) and a model was developed to predict possible effects in food systems.

Nisin is more effective in more acidic foods but *L. monocytogenes*, which has adapted to acidic conditions, becomes more tolerant of nisin (*34*). This tolerance, along with the development of nisin-resistant strains (*23*) and mutants (*27,29*) of *L. monocytogenes* may limit the effectiveness of nisin in some applications. One solution is the use of nisin in combination with another bacteriocin, e.g. leucocin F 10 (*28*) or with starter cultures of bacteria producing other antilisterial bacteriocins (*29*).

Recently, powders containing nisin and pediocin were produced from milk-based media and applied to food packaging materials (*25*). The bacteriocins did not diffuse through casings and packaging films and effectively inhibited listerial growth on meat surfaces.

In experiments with nisin used as a dip for meats, growth of *L. monocytogenes* on raw pork tenderloin (*11*), fresh ground pork (*27*), and cooked pork tenderloin (*10*) was inhibited. However, after a short time under aerobic conditions at 5°C, nisin-resistant listeriae started to grow on the pork. Modified atmosphere packaging provided an additional hurdle and margin of safety.

Nisin also inhibited the growth of *L. monocytogenes* on beef steaks (*1*) and cubes (*6,40*). Although vacuum packaging alone did not prevent listerial growth on steaks, nisin added to the meat before vacuum packaging effectively suppressed the growth of *L. monocytogenes* for 4 weeks at 4°C (*1*). Inhibition of listerial growth on beef cubes was greater at refrigeration temperatures but even at room temperature growth was delayed for one day (*6*). This may afford some protection during short periods of temperature abuse. EDTA does not enhance the antilisterial activity of nisin on beef (*40*).

Other experiments indicated that a rinse with nisin reduced populations of *L. monocytogenes* attached to turkey skin and growth was further inhibited during refrigerated storage (*22*).

Pediocin AcH. Pediocin has strong antilisterial effects in culture with a lower minimal inhibitory concentration (MIC) than nisin A or Z (*24*). However, in meat such as ground pork, this bacteriocin reduces *L. monocytogenes* populations by as much as 2 logs within 24 hours (*19*) but it loses its effectiveness over time apparently due to

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison
November 1999

its rapid degradation by meat proteases (*27*). Encapsulation of pediocin in liposomes or the addition of an emulsifier (Tween 80) increased its antilisterial effects in beef slurries (*7*). Pediocin can also be used in combination with other preservatives, such as diacetate, lactate and nitrite, to ensure greater inhibition of *L. monocytogenes* in turkey slurries (*31*). Pediocin and pediocin-producing cultures added to wiener exudates killed *L. monocytogenes* at both refrigeration and room temperatures (*38*). In addition, pediocin-producing bacteria, added as part of starter cultures for the production of chicken summer sausage, killed listeriae during fermentation (*3*).

One advantage of using pediocin in meats is its resistance to thermal degradation. It can be added to raw chicken and will retain its activity after the chicken is cooked (*13*). Pediocin-containing powders have been produced and applied to food packaging films that inhibit the growth of *L. monocytogenes* on the surface of meat (*25*).

Reuterin. Reuterin (produced by *Lactobacillus reuteri*) is a broad-spectrum antimicrobial agent which is water-soluble, effective over a wide pH range, and resistant to proteolytic and lipolytic enzymes. When added to the surface of cooked pork or mixed with ground pork, reuterin reduced populations of *L. monocytogenes* by 0.3 and 3.0 logs, respectively. Lactic acid enhanced the effectiveness of this bacteriocin (*9*).

Sakacin. Sakacin P, produced by *Lactobacillus sake* LTH 673, inhibits the growth of *Listeria ivanovii*, and this inhibition is increased by high NaCl concentrations and a low pH (*12*). Sakacin K, produced by *L. sake* CTC 494, inhibited the growth of *Listeria innocua* in raw minced pork, poultry breast meat, and cooked pork. The greatest reduction in listerial populations occurred in meats packaged in vacuum or modified atmospheres (*17*). *Lactobacillus sake* CTC 494 appears to be a very useful organism for sausage starter cultures because the temperature and pH conditions present during fermentation of dry sausages are ideal for sakacin K production (*21*).

REFERENCES

1. Avery SM, Buncic S. Antilisterial effects of a sorbate-nisin combination in vitro and on packaged beef at refrigeration temperature. J. Food Protect. 1997; 60(9):1075–1080.

2. Aymerich MT, Hugas M, Monfort JM. Bacteriocinogenic lactic acid bacteria associated with meat products. Food Sci. Technol. Int. 1998; 4(3):141–158.

3. Baccus-Taylor G, Glass KA, Luchansky JB, Maurer AJ. Fate of *Listeria monocytogenes* and pediococcal starter cultures during the manufacture of chicken summer sausage. Poultry Sci. 1993; 72(9):1772–1778.

4. Buncic S, Avery SM, Moorhead SM. Insufficient anti-listerial capacity of low inoculum *Lactobacillu*s cultures on long-term stored meats at 4°C. Int. J. Food Microbiol. 1997; 34(2):157–170.

5. Campos CA, Mazzotta, AS, Montville TJ. Inhibition of *Listeria monocytogenes* by *Carnobacterium piscicola* in vacuum-packaged cooked chicken at refrigeration temperatures. J. Food Safety 1997; 17(3):151–160.

6. Chung KT, Dickson JS, Crouse JD. Effects of nisin on growth of bacteria attached to meat. Appl. Environ. Microbiol. 1989; 55(6):1329–1333.

7. Degnan AJ, Buyong N, Luchansky JB. Antilisterial activity of pediocin AcH in model food systems in the presence of an emulsifier or encapsulated within liposomes. Int. J. Food Microbiol. 1993; 18(2):127–138.

8. De Martinis ECP, Franco BDGM. Inhibition of *Listeria monocytogenes* in a pork product by a *Lactobacillus sake* strain. Int. J. Food Microbiol. 1998; 42(1–2):119–126.

9. El-Ziney MG, van den Tempel T, Debevere J, Jakobsen M. Application of reuterin produced by *Lactobacillus reuteri* 12002 for meat decontamination and preservation. J. Food Protect. 1999; 62(3):257–261.

10. Fang TJ, Lin LW. Growth of *Listeria monocytogenes* and *Pseudomonas fragi* on cooked pork in a modified atmosphere packaging/nisin combination system. J. Food Protect. 1994; 57(6):479–485.

11. Fang TJ, Lin LW. Inactivation of *Listeria monocytogenes* on raw pork treated with modified atmosphere packaging and nisin. J. Food Drug Anal. 1994; 2(3):189–200.

12. Ganzle MG, Hertel C, Hammes WP. Antimicrobial activity of bacteriocin-producing cultures in meat products — modelling of the effect of pH, NaCl, and nitrite concentrations on the antimicrobial activity of sakacin p against *Listeria ivanovii* dsm20750. Fleischwirtschaft 1996; 76(4):409–412.

13. Goff JH, Bhunia AK, Johnson MG. Complete inhibition of low levels of *Listeria monocytogenes* on refrigerated chicken meat with pediocin AcH bound to heat-killed *Pediococcus acidilactici* cells. J. Food Protect. 1996; 59(11):1187–1192.

14. Holley RA, Doyon G, Fortin J, Rodrigue N, Carbonneau M. Post-process, packaging-induced fermentation of delicatessen meats. Food Res. Int. 1996; 29(1):35–48.

15. Hugas M. Bacteriocinogenic lactic acid bacteria for the biopreservation of meat and meat products. Meat Sci. 1998; 49(Suppl 1):S139–S150.

16. Hugas M, Neumeyer B, Pages F, Garriga M, Hammes WP. Antimicrobial activity of bacteriocin-producing cultures

Control of *Listeria* in Meat by Bacteriocins

in meat products. 2. Comparison of the antilisterial potential of bacteriocin-producing lactobacilli in fermenting sausages. Fleischwirtschaft 1996; 76(6):649–652.

17. Hugas M, Pages F, Garriga M, Monfort JM. Application of the bacteriocinogenic *Lactobacilus sakei* CTC494 to prevent growth of *Listeria* in fresh and cooked meat products packed with different atmospheres. Food Microbiol. 1998; 15(6):639–650.

18. Juven BJ, Barefoot SF, Pierson MD, McCaskill LH, Smith B. Growth and survival of *Listeria monocytogenes* in vacuum-packaged ground beef inoculated with *Lactobacillus alimentarius* FloraCarn l–2. J. Food Protect. 1998; 61(5):551–556.

19. Khojasteh A, Murano EA. Inability of heat stress to affect sensitivity of *Listeria monocytogenes* to pediocin in pork. J. Food Safety 1996; 16(3):201–208.

20. Leisner JJ, Greer GG, Stiles, ME. Control of beef spoilage by a sulfide-producing *Lactobacillus sake* strain with bacteriocinogenic *Leuconostoc gelidum* ual187 during anaerobic storage at 2°C. Appl. Environ. Microbiol. 1996; 62(7):2610–2614.

21. Leroy F, De Vuyst L. Temperature and pH conditions that prevail during fermentation of sausages are optimal for production of the antilisterial bacteriocin sakacin K. Appl. Environ. Microbiol. 1999; 65(3):974–981.

22. Mahadeo M, Tatini SR. The potential use of nisin to control *Listeria monocytogenes* in poultry. Lett. Appl. Microbiol. 1994; 18:323–326.

23. Mazzotta AS, Montville TJ. Nisin induces changes in membrane fatty acid composition of *Listeria monocytogenes* nisin-resistant strains at 10°C and 30°C. J. Appl. Microbiol. 1997; 82(1):32–38.

24. Meghrous J, Lacroix C, Simard RE. The effects on vegetative cells and spores of three bacteriocins from lactic acid bacteria. Food Microbiol. 1999; 16(2):105–114.

25. Ming XT, Weber GH, Ayres JW, Sandine WE. Bacteriocins applied to food packaging materials to inhibit *Listeria monocytogenes* on meats. J. Food Sci. 1997; 62(2):413–415.

26. Muriana PM. Bacteriocins for control of *Listeria* spp. in food. J. Food Protect. 1996; 59(Suppl S):54–63

27. Murray M, Richard JA. Comparative study of the antilisterial activity of nisin A and pediocin AcH in fresh ground pork stored aerobically at 5°C. J. Food Protect. 1997; 60(12):1534–1540.

28. Parente E, Giglio MA, Ricciardi A, Clementi F. The combined effect of nisin, leucocin F10, pH, NaCl and EDTA on the survival of *Listeria monocytogenes* in broth. Int. J. Food Microbiol. 1998; 40(1–2):65–75.

29. Schillinger U, Chung HS, Keppler K, Holzapfel WH. Use of bacteriocinogenic lactic acid bacteria to inhibit spontaneous nisin-resistant mutants of *Listeria monocytogenes* Scott A. J. Appl. Microbiol. 1998; 85(4):657–663.

30. Schillinger U, Kaya M, Lucke FK. Behaviour of *Listeria monocytogenes* in meat and its control by a bacteriocin-producing strain of *Lactobacillus sake*. J. Appl. Bacteriol. 1991; 70(6):473–478.

31. Schlyter JH, Glass KA, Loeffelholz J, Degnan AJ, Luchansky JB. The effects of diacetate with nitrite, lactate, or pediocin on the viability of *Listeria monocytogenes* in turkey slurries. Int. J. Food Microbiol. 1993; 19(4):271–281.

32. Schöbitz R, Zaror T, Leon O, Costa M. A bacteriocin from *Carnobacterium piscicola* for the control of *Listeria monocytogenes* in vacuum-packaged meat. Food Microbiol. 1999; 16(3):249–255.

33. ter Steeg PF, Hellemons JC, Kok AE. Synergistic actions of nisin, sublethal ultrahigh pressure, and reduced temperature on bacteria and yeast. Appl. Environ. Microbiol. 1999; 65(9):4148–4154.

34. van Schaik W, Gahan CGM, Hill C. Acid-adapted *Listeria monocytogenes* displays enhanced tolerance against the lantibiotics nisin and lacticin 3147. J. Food Protect. 1999; 62(5):536–539.

35. Vignolo G, Fadda S, de Kairuz MN, Holgado APD, Oliver, G. Effects of curing additives on the control of *Listeria monocytogenes* by lactocin 705 in meat slurry. Food Microbiol. 1998; 15(3):259–264.

36. Vignolo G, Fadda S, de Kairuz MN, Holgado AAPD, Oliver G. Control of *Listeria monocytogenes* in ground beef by lactocin 705, a bacteriocin produced by *Lactobacillus casei* CRL 705. Int. J. Food Microbiol. 1996; 29(2–3):397–402.

37. Villani F, Sannino L, Moschetti G, Mauriello G, Pepe O, Amodio-Cocchieri R, Coppola S. Partial characterization of an antagonistic substance produced by *Staphylococcus xylosus* 1E and determination of the effectiveness of the producer strain to inhibit *Listeria monocytogenes* in Italian sausages. Food Microbiol. 1997; 14(6):555–566.

38. Yousef AE, Luchansky JB, Degnan AJ, Doyle MP. Behavior of *Listeria monocytogenes* in wiener exudates in the presence of *Pediococcus acidilactici* H or pediocin AcH during storage at 4 or 25°C. Appl. Environ. Microbiol. 1991; 57(5):1461–1467.

39. Yuste J, Mormur M, Capellas M, Guamis B, Pla R. Microbiological quality of mechanically recovered poultry meat treated with high hydrostatic pressure and nisin. Food Microbiol. 1998; 15(4):407–414.

40. Zhang SS, Mustapha A. Reduction of *Listeria monocytogenes* and *Escherichia coli* O157:H7 numbers on vacuum-packaged fresh beef treated with nisin or nisin combined with EDTA. J. Food Protect. 1999; 62(10):1123–1127.

USE OF THERMAL PROCESSES TO CONTROL *LISTERIA* IN MEAT

Heat resistance of *Listeria monocytogenes* depends upon many factors including characteristics of different strains and serovars (*2,32,44*). Conditions known to affect the susceptibility of *L. monocytogenes* to thermal treatments include stage in the growth cycle, temperature during growth, and exposure to other stresses. Cells in stationary phase (*31*), those grown at higher temperatures (19 or 37°C) (*2,27*), and those previously exposed to stresses such as acid, ethanol, and hydrogen peroxide (*31*) are generally more resistant to thermal treatments. Thermotolerance is increased significantly after heat shock (30 min exposure to 48°C) in cells grown at 4°C (*26*) and tends to increase in cells grown at higher temperatures (*4,5,13,14*). Ranges of D values measured for *L. monocytogenes* in various types of meat are presented in a table at the end of this report.

Beef. In raw ground beef, higher concentrations of fat (30.5%) appear to protect *L. monocytogenes* from heat while higher concentrations of lactate enhance bacterial destruction by heat (*12*). In the production of beef jerky, *L. monocytogenes* populations are reduced during heating and marination and become undetectable after a 10-hour drying period (*23*). For production of microwave-ready roast beef, cooking in a bag was twice as effective as without the bag since *L. monocytogenes* could survive on beef surfaces which had been cooked for up to 45 min to a temperature of 62.8°C (*47*). Heating of beef loin chunks for 16 min at 85°C reduced *L. monocytogenes* populations by as much as 4 logs. However, some cells survived and might be able to grow under appropriate conditions (*9*). In a process simulating sous vide preparation of cooked beef, with slow heating, *L. monocytogenes* was killed as efficiently by the slow heating process as by faster heating. The reason for this difference from tests in pork (*30,40*) appears to be the low pH of 5.64 of the beef (*19*).

Pork. *Listeria monocytogenes* is more heat resistant when mixed with raw ground pork than when suspended in broth medium (*40*). Addition of soy hulls to ground pork further protects listeriae from heat (*38*). When pork inoculated with *L. monocytogenes* is heated slowly, the thermal tolerance of these bacteria is much greater as compared to bacteria in pork heated rapidly (*30,40*).

Cured meats. Investigations with beaker sausage demonstrated that heating the sausage to an internal temperature of 62.8°C was required to completely inactivate *L. monocytogenes* (*17*). Heating pepperoni at 51.7°C for 4 hours after drying destroyed listeriae but heating before drying was insufficient to eliminate the bacteria (*17*).

Curing agents (usually a mixture of sodium chloride, sodium nitrite/nitrate, dextrose, etc.) protect *L. monocytogenes* in various types of sausage, ham, bologna and other cured meats from thermal destruction (*13,29,32,43,48,49*). When curing ingredients were con-

D value ranges (min) for thermal inactivation of *L. monocytogenes* in different meats

Meat	D values at 60°C	Reference number(s)
ground beef – raw	0.24 – 12.53*	3,11,25,32
ground beef – cooked	6.27 – 8.32	15
ground chicken – raw	5.6 – 8.7	32
ground chicken – cooked	5.02 – 5.29	15
ground pork – raw	4.3 – 9.2 (62°C)	30
ham	1.82	5
sausage	7.3 – 9.13	2,40
sous vide beef	6.4 – 7.1	19
roast beef	1.625	18
beaker sausage	9.13	43

*Variability related to differences in strains, pH, log vs stationary phase cells, heating rate.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

sidered separately, all except sodium nitrite and sodium erythorbate enhanced listerial thermotolerance in ground pork (15% fat) (*48*). Addition of k-carrageenan to cured ground pork lessened the protective effects of curing salts (*49*).

While many thermal processing treatments are very effective in killing foodborne pathogens, high temperatures or prolonged heating may alter some sensory characteristics of foods. Therefore, research is underway to determine appropriate combinations of heat and high pressure treatments (*1,28,36,45*), irradiation, bacteriocins, or other antimicrobials (*46*) to produce safe and more organoleptically acceptable foods.

REFERENCES

1. Alpas H, Kalchayanand N, Bozoglu F, Ray B. Interaction of pressure, time and temperature of pressurization on viability loss of *Listeria innocua*. World J. Microbiol. Biotechnol. 1998; 14:251–253.

2. Bhaduri S, Smith PW, Palumbo SA, Turner-Jones CO, Smith JL, Marmer BS, Buchanan RL, Zaika LL, Williams AC. Thermal destruction of *Listeria monocytogenes* in liver sausage slurry. Food Microbiol. 1991; 8:75–78.

3. Boyle DL, Sofos JN, Schmidt GR. Thermal destruction of *Listeria monocytogenes* in a meat slurry and in ground beef. J. Food Sci. 1990; 55:327–329.

4. Bunning VK, Crawford RG, Tierney JT, Peeler JT. Thermotolerance of *Listeria monocytogenes* and *Salmonella typhimurium* after sublethal heat shock. Appl. Environ. Microbiol. 1990; 56:3216–3219.

5. Carlier V, Augustin JC, Rozier J. Heat resistance of *Listeria monocytogenes* (Phagovar 2389/2425/3274/2671/47/108/340): D- and z-values in ham. J. Food Prot. 1996; 59:588–591.

6. Carlier V, Augustin JC, Rozier J. Destruction of *Listeria monocytogenes* during a ham cooking process. J. Food Prot. 1996; 59:592–595.

7. Carpenter SL, Harrison MA. Fate of small populations of *Listeria monocytogenes* on poultry processed using moist heat. J. Food Prot. 1989; 52:768–770.

8. Carpenter SL, Harrison MA. Survival of *Listeria monocytogenes* on processed poultry. J. Food Sci. 1989; 54:556–557.

9. Cooksey DK, Klein BP, McKeith FK, Blaschek HP. Reduction of *Listeria monocytogenes* in precooked vacuum-packaged beef using postpackaging pasteurization. J. Food Prot. 1993; 56:1034–1038.

10. Dealler S, Rotowa N, Lacey R. Microwave reheating of convenience meals. Br. Food J. 1990; 92:19–21.

11. Doherty AM, McMahon CMM, Sheridan JJ, Blair IS, McDowell DA, Hegarty T. Thermal resistance of *Yersinia enterocolitica* and *Listeria monocytogenes* in meat and potato substrates. J. Food Safety 1998; 18:69–83.

12. Fain AR Jr, Line JE, Moran AB, Martin LM, Lechowich RV, Carosella JM, Brown WL. Lethality of heat to *Listeria monocytogenes* Scott A: D-value and z-value determinations in ground beef and turkey. J. Food Prot. 1991; 54:756–761.

13. Farber JM. Thermal resistance of *Listeria monocytogenes* in foods. Int. J. Food Microbiol. 1989; 8:285–291.

14. Farber JM, Brown BE. Effect of prior heat shock on heat resistance of *Listeria monocytogenes* in meat. Appl. Environ. Microbiol. 1990; 56:1584–1587.

15. Gaze JE, Brown GD, Gaskell DE, Banks JG. Heat resistance of *Listeria monocytogenes* in homogenates of chicken, beef steak and carrot. Food Microbiol. 1989; 6:251–259.

16. George SM, Richardson LCC, Peck MW. Predictive models of the effect of temperature, pH and acetic and lactic acids on the growth of *Listeria monocytogenes*. Int. J. Food Microbiol. 1996; 32:73–90.

17. Glass KA, Doyle MP. Fate and thermal inactivation of *Listeria monocytogenes* in beaker sausage and pepperoni. J. Food Prot. 1989; 52:226–231.

18. Grant IR, Patterson MF. Combined effect of gamma radiation and heating on the destruction of *Listeria monocytogenes* and *Salmonella typhimurium* in cook-chill roast beef and gravy. Int. J. Food Microbiol. 1995; 27:117–128.

19. Hansen TB, Knøchel S. Thermal inactivation of *Listeria monocytogenes* during rapid and slow heating in sous vide cooked beef. Lett. Appl. Microbiol. 1996; 22:425–428.

20. Hardin MD, Williams SE, Harrison MA. Survival of *Listeria monocytogenes* in postpasteurized precooked beef roasts. J. Food Prot. 1993; 56:655–660.

21. Harrison MA, Carpenter SL. Survival of large populations of *Listeria monocytogenes* on chicken breasts processed using moist heat. J. Food Prot. 1989; 52:376–378.

22. Harrison MA, Carpenter SL. Survival of *Listeria monocytogenes* on microwave cooked poultry. Food Microbiol. 1989; 6:153–157.

23. Harrison JA, Harrison MA. Fate of *Escherichia coli* O157:H7, *Listeria monocytogenes, and Salmonella typhimurium* during preparation and storage of beef jerky. J. Food Prot. 1996; 59:1336–1338.

24. Huang IPD, Yousef AE, Marth EH, Matthews ME. Thermal inactivation of *Listeria monocytogenes* in chicken gravy. J. Food Prot. 1992; 55:492–496.

25. Jørgensen F, Hansen TB, Knochel S. Heat shock-induced thermotolerance in *Listeria monocytogenes* 13-249 is dependent on growth phase, pH and lactic acid. Food Microbiol. 1999; 16:185–194.

26. Jørgensen F, Panaretou B, Stephens PJ, Knøchel S. Effect of pre- and post-heat shock temperature on the persistence of thermotolerance and heat shock-induced proteins in *Listeria monocytogenes*. J. Appl. Bacteriol. 1996; 80:216–224.

27. Juneja VK, Eblen BS. Predictive thermal inactivation model for *Listeria monocytogenes* with temperature, pH,

NaCl, and sodium pyrophosphate as controlling factors. J. Food Prot. 1999; 62:986–993.

28. Kalchayanand N, Sikes A, Dunne CP, Ray B. Interaction of hydrostatic pressure, time and temperature of pressurization and pediocin AcH on inactivation of foodborne bacteria. J. Food Prot. 1998; 61:425–431.

29. Kamat AS, Nair PM. Identification of *Listeria innocua* as a biological indicator for inactivation of *L. monocytogenes* by some meat processing treatments. Food Sci. Technol. 1996; 29:714–720.

30. Kim KT, Murano EA, Olson DG. Heating and storage conditions affect survival and recovery of *Listeria monocytogenes* in ground pork. J. Food Sci. 1994; 59:30–32, 39.

31. Lou Y, Yousef AE. Resistance of *Listeria monocytogenes* to heat after adaptation to environmental stresses. J. Food Prot. 1996; 59:465–471.

32. Mackey BM, Pritchet C, Norris A, Mead GC. Heat resistance of *Listeria*: strain differences and effects of meat type and curing salts. Lett. Appl. Microbiol. 1990; 10:251–255.

33. McMahon CMM, Doherty AM, Sheridan JJ, Blair IS, McDowell DA, Hegarty T. Synergistic effect of heat and sodium lactate on the thermal resistance of *Yersinia enterocolitica* and *Listeria monocytogenes* in minced beef. Lett. Appl. Microbiol. 1999; 28:340–344.

34. Miles CA, Mackey BM. A mathematical analysis of microbial inactivation at linearly rising temperatures: calculation of the temperature rise needed to kill *Listeria monocytogenes* in different foods and methods for dynamic measurements of D and z values. J. Appl. Bacteriol. 1994; 77:14–20.

35. Morgan AI, Goldberg N, Radewonuk ER, Scullen OJ. Surface pasteurization of raw poultry meat by steam. Food Sci. Technol. 1996; 29:447–451.

36. Murano EA, Murano PS, Brennan RE, Shenoy K, Moreira RG. Application of high hydrostatic pressure to eliminate *Listeria monocytogenes* from fresh pork sausage. J. Food Prot. 1999; 62:480–483.

37. Murphy RY, Marks BP, Johnson ER, Johnson MG. Inactivation of *Salmonella* and *Listeria* in ground chicken breast meat during thermal processing. J. Food Prot. 1999; 62:980–985.

38. Ollinger-Snyder P, El-Gazzar F, Matthews ME, Marth EH, Unklesbay N. Thermal destruction of *Listeria monocytogenes* in ground pork prepared with and without soy hulls. J. Food Prot. 1995; 58:573–576.

39. Palumbo SA, Smith JL, Marmer BS, Zaika LL, Bhaduri S, Turner-Jones C, Williams AC. Thermal destruction of *Listeria monocytogenes* during liver sausage processing. Food Microbiol. 1993; 10:243–247.

40. Quintavalla S, Campanini M. Effect of rising temperature on the heat resistance of *Listeria monocytogenes* in meat emulsion. Lett. Appl. Microbiol. 1991; 12:184–187.

41. Roering AM, Wierzba RK, Ihnot AM, Luchansky JB. Pasteurization of vacuum-sealed packages of summer sausage inoculated with *Listeria monocytogenes*. J. Food Safety 1988; 18:49–56.

42. Samelis J, Kakouri A, Georgiadou KG, Metaxopoulos J. Evaluation of the extent and type of bacterial contamination at different stages of processing of cooked ham. J. Appl. Microbiol. 1998; 84:649–660.

43. Schoeni JL, Brunner K, Doyle MP. Rates of thermal inactivation of *Listeria monocytogenes* in beef and fermented beaker sausage. J. Food Prot. 1991; 54:334–337.

44. Sörqvist S. Heat resistance of different serovars of *Listeria monocytogenes*. J. Appl. Bacteriol. 1994; 76:383–388.

45. Stewart CM, Jewett FF, Dunne CP DG. Hoover. Effect of concurrent high hydrostatic pressure, acidity and heat on the injury and destruction of *Listeria monocytogenes*. J. Food Safety 1997; 17:23–36.

46. Ueckert JE, ter Steeg PF, Coote PJ. Synergistic antibacterial action of heat in combination with nisin and magainin II amide. J. Appl. Microbiol. 1998; 85:487–494.

47. Unda JR, Molins RA, Walker HW. *Clostridium sporogenes* and *Listeria monocytogenes*: survival and inhibition in microwave-ready beef roasts containing selected antimicrobials. J. Food Sci. 1991; 56:198–205,219.

48. Yen LC, Sofos JN, Schmidt GR. Effect of meat-curing ingredients on thermal destruction of *Listeria monocytogenes* in ground pork. J. Food Prot. 1991; 54:408–412.

49. Yen LC, Sofos JN, Schmidt GR. Destruction of *Listeria monocytogenes* by heat in ground pork formulated with kappa-carrageenan, sodium lactate and the algin/calcium meat binder. Food Microbiol. 1992; 9:223–230.

50. Zaika LL, Palumbo SA, Smith JL, DelCorral F, Bhaduri S, Jones CO, Kim AH. Destruction of *Listeria monocytogenes* during frankfurter processing. J. Food Prot. 1990; 53:18–21.

USE OF IRRADIATION TO CONTROL *LISTERIA* IN MEAT

Irradiation can damage and destroy most foodborne bacteria, including *Listeria monocytogenes*. [See Fu et al. (*6*) for a recent review.] Irradiation dosage, expressed in kiloGrays (kGy), is a function of the energy of the radiation source and the time of exposure. Effectiveness of a given radiation dose varies depending on the density, antioxidant levels, moisture, and other components or characteristics of the foods. External factors, such as temperature, the presence or absence of oxygen, and subsequent storage conditions also influence the effectiveness of radiation. A split dose application of irradiation increased the radiosensitivity of *L. monocytogenes* to irradiation under some conditions (*1*).

Different isolates of *L. monocytogenes* exhibit some variation in resistance to irradiation. Under similar experimental conditions, the range in D_{10} values in: (a) culture media was 0.28–0.34 kGy (*11*); (b) mechanically deboned chicken meat was 0.41–0.53 kGy (*11*); (c) minced raw chicken was 0.48–0.54 kGy (*15*); (d) ground beef was 0.5–1.0 kGy (*3*); and (e) ground pork was 0.42–0.64 kGy (*19*). *Listeria innocua*, a nonpathogenic species, is similar to *L. monocytogenes* in its sensitivity to irradiation and so may be used for the safe evaluation of irradiation processes for different meats (*13*).

In nearly all experiments, *Salmonella* and *Listeria* proved to be more resistant to irradiation than *E. coli*, *Arcobacter*, *Campylobacter*, *Yersinia*, and *Staphylococcus* (*5,6,7,8,14, 22*). *Listeria* and *Salmonella* appear to have a similar susceptibility to irradiation; in some experiments, *L. monocytogenes* has a larger D_{10} value while in other cases, *Salmonella* appears to be more resistant (*4,6,7,8,9,22*).

Irradiation of *L. monocytogenes* in laboratory media offers some useful preliminary information but *L. monocytogenes* is significantly more resistant to irradiation in meats than in culture media (*2,3,10,11,12, 13,15*). However, neither the fat content of the meat (*14*) nor the source (beef, chicken, lamb, pork, turkey breast, turkey leg) of raw meat (*12,22*) had a significant effect on D values for irradiation.

Factors that do affect the effectiveness of a radiation dose in meat include cooking, concentration of bacteria in the meat, and temperature during irradiation. *Listeria monocytogenes* added to raw turkey nuggets was more susceptible to irradiation than that added to cooked turkey nuggets (*23*). At lower temperatures, the radiation resistance of *L. monocytogenes* increased (*2,12,20*). With larger concentrations of *L. monocytogenes* in solution or on meat, larger doses of radiation are required to destroy the cells (*2,16*). Therefore, if food is highly contaminated, the usual radiation dose may not kill all the *L. monocytogenes* and, as several researchers reminded us, *L. monocytogenes* can grow in the cold and surviving and damaged cells may begin to multiply if the irradiated meat is stored under refrigeration (*10,24*).

Heat treatments as in sous vide processing (*9,17, 18*) and modified atmosphere packaging (*7,21,24*) have been found to enhance the safety of irradiated foods. In addition, salt, nitrites, and other compounds added to preserved meats may increase the effectiveness of a radiation dose: *L. monocytogenes* is more radiation-resistant in uncured pork than in ham (*4*). These additives may act by amplifying the kill by irradiation or by preventing the repair and growth of damaged, surviving cells. However, there has been very little published research on the effects of irradiation on cured and processed meats.

Some recommended doses of irradiation include: (a) 3 kGy for elimination of 10^3 cells *L. monocytogenes* per g in air-packed frozen chicken (*12*); (b) 2.5 kGy to kill $10^{4.1}$ *L. monocytogenes* per g in ground beef (*14*); and (c) 2 kGy to destroy 10^4 *L. monocytogenes* in mechanically deboned chicken meat at 2–4ºC (*11*).

Food processors should be aware that various food additives and changes in processing parameters may affect the effectiveness of a radiation dose and that any surviving *Listeria* may grow to dangerous levels during storage at refrigeration temperatures if some other hurdle(s) to growth are not present. In addition, only a few types of plastic wraps and packaging are approved for use in irradiating packaged foods.

Irradiation has been approved by the FDA (*25,26*) for the purpose of microbial disinfestation of:

product	to a limit of
fresh or frozen uncooked poultry	<3.0 kGy
pork carcasses and meat (for *Trichinella*)	<1.0 kGy
packaged meat for NASA flights	<44 kGy
fresh red meat	4.5 kGy
frozen red meat	7 kGy

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

Control of *Listeria* in Meat by Irradiation

Irradiation of red meat (not including processed ready-to-eat meats) was approved by the FDA in December 1997, and the recommended procedures for irradiating meat have been published by the USDA in the *Federal Register* (*26*). Since the period for comments on these procedures has been extended, the final rules have not been published as yet (July 1, 1999).

A number of individual European countries have regulations in place permitting (or in some cases prohibiting) irradiation of foods under specified conditions. The European Community is at this time working to establish a common set of guidelines.

REFERENCES

1. Andrews L, Grodner RM. Radiosensitivity of *Listeria monocytogenes* using split dose application of gamma irradiation. J. Food Protect. 1997; 60(3):262–266.
2. Andrews LS, Marshall DL, Grodner RM. Radiosensitivity of *Listeria monocytogenes* at various temperatures and cell concentrations. J. Food Protect. 1995; 58(7):748–751.
3. El Shenawy MA, Yousef AE, Marth EH. Radiation sensitivity of *Listeria monocytogenes* in broth or in raw ground beef. Lebensm. Wiss. Technol. 1989; 22(6):387–390.
4. Farkas J. Irradiation as a method for decontaminating food – a review. Int. J. Food Microbiol. 1998; 44(3):189–204.
5. Fu AH, Sebranek JG, Murano EA. Survival of *Listeria monocytogenes*,*Yersinia enterocolitica* and *Escherichia col*i O157:H7 and quality changes after irradiation of beef steaks and ground beef. J. Food Sci. 1995; 60(5):972–977.
6. Fu AH, Sebranek JG, Murano EA. Survival of *Listeria monocytogenes* and *Salmonella typhimurium* and quality attributes of cooked pork chops and cured ham after irradiation. J. Food Sci. 1995; 60(5):1001–1005, 1008.
7. Grant IR, Patterson MF. Effect of irradiation and modified atmosphere packaging on the microbiological safety of minced pork stored under temperature abuse conditions. Int. J. Food Sci. Technol. 1991; 26(5):521–533.
8. Grant IR, Patterson MF. Sensitivity of foodborne pathogens to irradiation in the components of a chilled ready meal. Food Microbiol. 1992; 9(2):95–103.
9. Grant IR, Patterson MF. Combined effect of gamma radiation and heating on the destruction of *Listeria monocytogenes* and *Salmonella typhimurium* in cook-chill roast beef and gravy. Int. J. Food Microbiol. 1995; 27(2/3):117–128.
10. Gursel B, Gurakan GC. Effects of gamma irradiation on the survival of *Listeria monocytogenes* and on its growth at refrigeration temperature in poultry and red meat. Poultry Sci. 1997; 76(12):1661–1664.
11. Huhtanen CN, Jenkins RK, Thayer DW. Gamma radiation sensitivity of *Listeria monocytogenes*. J. Food Protect. 1989; 52(9):610–613.
12. Kamat AS, Nair MP. Gamma irradiation as a means to eliminate *Listeria monocytogenes* from frozen chicken meat. J. Sci. Food Agric. 1995; 69(4):415–422.
13. Kamat AS, Nair PM. Identification of *Listeria innocua* as a biological indicator for inactivation of *L. monocytogenes* by some meat processing treatments. Food Sci. Technol.–Lebensm.-Wiss. Technol. 1996; 29(8):714–720.
14. Monk JD, Clavero MRS, Beuchat LR, Doyle MP, Brackett RE. Irradiation inactivation of *Listeria monocytogenes* and *Staphylococcus aureus* in low- and high-fat, frozen and refrigerated ground beef. J. Food Protect. 1994; 57(11):969–974.
15. Patterson M. Sensitivity of *Listeria monocytogenes* to irradiation on poultry meat and in phosphate-buffered saline. Lett. Appl. Microbiol. 1989; 8(5):181–184.
16. Patterson MF, Damoglou AP, Buick RK. Effects of irradiation dose and storage temperature on the growth of *Listeria monocytogenes* on poultry meat. Food Microbiol. 1993; 10(3):197–203.
17. Shamsuzzaman K, Chuaqui Offermanns N, Lucht L, McDougall T, Borsa J. Microbiological and other characteristics of chicken breast meat following electron-beam and sous-vide treatments. J. Food Protect. 1992; 55(7):528–533.
18. Shamsuzzaman K, Lucht L, Chuaqui Offermanns N. Effects of combined electron-beam irradiation and sous-vide treatments on microbiological and other qualities of chicken breast meat. J. Food Protect. 1995; 58(5):497–501.
19. Tarté RR, Murano EA, Olson DG. Survival and injury of *Listeria monocytogenes*, *Listeria innocua* and *Listeria ivanovii* in ground pork following electron beam irradiation. J. Food Protect. 1996; 59(6):596–600.
20. Thayer DW, Boyd G. Radiation sensitivity of *Listeria monocytogenes* on beef as affected by temperature. J. Food Sci. 1995; 60(2):237–240.
21. Thayer DW, Boyd G. Irradiation and modified atmosphere packaging for the control of *Listeria monocytogenes* on turkey meat. J. Food Protect. 1999; 62(10):1136–1142.
22. Thayer DW, Boyd G, Fox JB Jr, Lakritz L, Hampson JW. Variations in radiation sensitivity of foodborne pathogens associated with the suspending meat. J. Food Sci. 1995; 60(1):63–67.
23. Thayer DW, Boyd G, Kim A, Fox JB, Farrell HM. Fate of gamma-irradiated *Listeria monocytogenes* during refrigerated storage on raw or cooked turkey breast meat. J. Food Protect. 1998; 61(8):979–987.
24. Varabioff Y, Mitchell GE, Nottingham SM. Effects of irradiation on bacterial load and *Listeria monocytogenes* in raw chicken. J. Food Protect. 1992; 55(5):389–391.
25. Food and Drug Administration. Irradiation in the Production, Processing and Handling of Food. Federal Register, vol. 62 (232) Dec. 3, 1997.
26. Food Safety and Inspection Service. Irradiation of Meat and Meat Products. Federal Register 64(36), Feb. 24, 1999.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

USE OF MODIFIED ATMOSPHERE PACKAGING TO CONTROL *LISTERIA* IN MEAT

Packaging of meats in modified atmospheres (MAP) containing low oxygen and/or high carbon dioxide levels can suppress the growth of foodborne pathogens as well as extend shelf life and preserve food quality. Several review papers discuss the advantages and disadvantages of various MAP systems with respect to the gases used, types of foods and packaging materials (*8,13,30*), effects on *Listeria monocytogenes* (*7,30*), and effectiveness of the combined use of MAP and irradiation (*23*). In addition, models have been developed to predict the growth of *L. monocytogenes* in culture media containing: (a) different concentrations of carbon dioxide (0–100%) and sodium chloride (0.5–8%) at pH 4.5–7.0 and 4–20°C (*15*); (b) carbon dioxide (10–90%) at pH 5.5–6.5 and 4–10°C (*12*); and (c) anaerobic nitrogen atmosphere with sodium chloride (0.5–4.5%) and sodium nitrite 50–1000 µg/ml at pH 6–7.5 and 5–37°C (*4*). Predicted listerial growth rates from one model were in good agreement with observed growth in chicken nuggets and raw and cooked beef (*15*). However, growth rates of *L. monocytogenes* on raw chicken were greater and on raw pork were much greater than those predicted by the model.

Results of numerous studies on the efficacy of different MAP systems in suppressing the growth of *L. monocytogenes* on different meats have been published in the past decade. However, data are not always consistent. This may result from variations in fat content and acidity of foods, storage temperatures, and the presence of other preservatives. MAP containing high levels of CO_2 effectively inhibit growth of *L. monocytogenes*, particularly at low temperatures. However, *L. monocytogenes* does grow in the absence of oxygen and has been observed to multiply on vacuum packaged meat at pH > 6. One general concern about MAP is that some atmospheres may inhibit spoilage bacteria but not significantly suppress *L. monocytogenes* or *Clostridium botulinum.* Therefore, after an extended period of refrigerated storage, the meat may appear to be unspoiled and safe to eat, but in fact it harbors high levels of these pathogens (*7,30*).

A brief summary of recent experimental results follows. Parameters that appeared to affect results are noted but original papers should be consulted for full experimental details.

Raw poultry. Storage temperature and carbon dioxide and oxygen levels in MAP significantly affect growth of *L. monocytogenes* on raw minced chicken (*33*), minced turkey (*32*), and turkey slices (*24*). An atmosphere containing 75% CO_2 inhibited growth at 4, 10, and 27°C but the addition of just 5% oxygen allowed growth at all of these temperatures (*33*). However, the presence of 60 or 80% oxygen prevented growth of *L. monocytogenes* at 1°C (*24*). Although irradiation (2.5 kGy) of ground turkey drastically reduced numbers of *L. monocytogenes*, surviving cells were able to grow at 7°C under atmospheres containing no oxygen and ≤ 64% CO_2 (*32*).

A lactate buffer, pH 3.0, combined with an atmosphere of 90% CO_2 inhibited growth of *L. monocytogenes* on chicken legs for nearly two weeks (*34*). Lactate by itself suppressed growth for about a week while the MAP alone suppressed growth for 2–4 days.

Cooked poultry. Temperature was also very important in limiting the growth of *L. monocytogenes* on cooked chicken breast (*4,6*), precooked chicken nuggets (*26,27*), chicken loaves (*18*), poultry cuts (*4*), and turkey roll slices (*14*). Despite vacuum packaging or atmospheres containing as much as 80% CO_2 and no oxygen, *L. monocytogenes* was able to grow on cooked poultry at temperatures between 6.5 and 11°C (*3,4,6,18,26,27*). At lower temperatures (6.5–7°C), MAP and the presence of lactate slowed the growth of *L. monocytogenes* somewhat even though they were not able to completely inhibit it (*3,6,18*). At 4°C, 70% CO_2 levels and vacuum packaging did suppress the growth of *L. monocytogenes* for 28 days in turkey roll slices (*14*) and chicken breast (*6*).

Raw pork. A study of the incidence of contaminated pork loins and Boston butts packaged in MAP revealed that very few butts were contaminated with *L. monocytogenes* while loins packaged under vacuum or in an atmosphere of 66% oxygen, 8% nitrogen, and 26% CO_2 had fewer contaminants than those packaged in air (*29*). Vacuum packaging did not prevent the growth of *L. monocytogenes* on hot or cold packed pork loin (*22*) or pork chops (*25*). Neither did vacuum packaging or a modified atmosphere (25% CO_2 : 75% nitrogen) prevent the growth, in ground pork, of listeriae injured by heat (*20*) or irradiation (*16*).

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

Control of *Listeria* in Meat by Modified Atmosphere Packaging

At 4°C, an atmosphere of 100% CO_2 did inhibit the growth of *L. monocytogenes* on raw pork tenderloin (*11*). Addition of nisin to pork tenderloin significantly suppressed growth of listeriae under both air and MAP (100% CO_2 and 80% CO_2 : 20% air) at both 4 and 20°C (*11*).

Cooked pork. *Listeria monocytogenes,* inoculated along with *Pseudomonas fragi,* on cooked pork tenderloin grew as well under modified atmospheres (100% CO_2 and 80% CO_2 : 20% air) as in air at both 4 and 20°C (*10*). Nisin solutions, used as 20 min dips for pork, prevented growth of *L. monocytogenes* under air and MAP at both temperatures.

Raw beef. Saturated carbon dioxide packaging but not vacuum packaging suppressed the growth of *L. monocytogenes* on beef steaks stored at 5 and 10°C for 3–6 weeks (*2*). Further experiments demonstrated that when contaminated steaks, which had been stored under a saturated carbon dioxide atmosphere at 1.5°C, were removed from storage and kept at 12°C (gross temperature abuse), *L. monocytogenes* still failed to grow or grew extremely slowly (*1*).

Although vacuum packaging alone was insufficient to prevent listerial growth in ground beef stored at 4°C for 9 weeks, the addition of *Lactobacillus alimentarius* L-2 to the beef caused about a 2 log decline in final numbers of *L. monocytogenes* (*19*). Since these lactobacilli do not produce bacteriocins, their inhibition is believed to be due to the production of lactic acid.

Cooked beef. Vacuum packaging of roast beef slices failed to prevent growth of *L. monocytogenes* at –1.5°C (*17*) or 3°C (*17,28*). A saturated carbon dioxide atmosphere caused *L. monocytogenes* populations to decline at –1.5°C and lengthened the lag phase at 3°C so that by the time *L. monocytogenes* grew the meat already appeared spoiled (*17*).

Cured meats. Tests with atmospheres containing 20, 30, 50 or 80% CO_2 demonstrated that only the highest carbon dioxide level was sufficient to inhibit growth of *L. monocytogenes* on frankfurters at both 4.7 and 10°C (*21*). An atmosphere with 50% carbon dioxide inhibited listerial growth only at the lower temperature. Neither vacuum packaging nor an atmosphere with 30% CO_2 : 70% nitrogen inhibited listerial growth on ham or lunch meat at 7°C (*4*).

Other uncured meats. Experiments with raw lamb pieces and mince demonstrated that listerial growth at 5°C was suppressed by an atmosphere of 100% CO_2 but not by atmospheres of 50% CO_2 : 50% nitrogen or 80% oxygen : 20% CO_2. Vacuum packaging was effective in preventing growth of listeriae in lamb mince but not in pieces of lamb (*31*). An atmosphere of 80% oxygen : 20% CO_2 was insufficient to prevent the growth of *L. monocytogenes* in lamb meat juice at 4°C (*9*).

REFERENCES

1. Avery SM, Rogers AR, Bell RG. Continued inhibitory effect of carbon dioxide packaging on *Listeria monocytogenes* and other microorganisms on normal pH beef during abusive retail display. Int. J. Food Sci. Technol. 1995; 30(6):725–735.

2. Avery SM, Hudson JA, Penney N. Inhibition of *Listeria monocytogenes* on normal ultimate pH beef (pH 5.3–5.5) at abusive storage temperatures by saturated carbon dioxide controlled atmosphere packaging. J. Food Protect. 1994; 57(4):331–333, 336.

3. Barakat RK, Harris LJ. Growth of *Listeria monocytogenes* and *Yersinia enterocolitica* on cooked modified-atmosphere-packaged poultry in the presence and absence of a naturally occurring microbiota. Appl. Environ. Microbiol. 1999; 65(1):342–345.

4. Beumer RR, Tegiffel MC, Deboer E, Rombouts FM. Growth of *Listeria monocytogenes* on sliced cooked meat products. Food Microbiol. 1996; 13(4):333–340.

5. Buchanan RL, Stahl HG, Whiting RC. Effects and interactions of temperature, pH, atmosphere, sodium chloride, and sodium nitrite on the growth of *Listeria monocytogenes*. J. Food Protect. 1989; 52(12):844–851.

6. Carpenter SL, Harrison MA. Survival of *Listeria monocytogenes* on processed poultry. J. Food Sci.1989; 54(3):556–557.

7. Defernando GDG, Nychas GJE, Peck MW, Ordonez JA. Growth survival of psychrotrophic pathogens on meat packaged under modified atmospheres. Int. J. Food Microbiol.1995; 28(2):221–231.

8. Devlieghere F, Debevere J, Vanimpe J. Concentration of carbon dioxide in the water-phase as a parameter to model the effect of a modified atmosphere on microorganisms. Int. J. Food Microbiol. 1998; 43(1–2):105–113.

9. Drosinos EH, Board RG. Growth of *Listeria monocytogenes* in meat juice under a modified atmosphere at 4ºC with or without members of a microbial association from chilled lamb under a modified atmosphere. Lett. Appl. Microbiol. 1994; 19(3):134–137.

10. Fang TJ, Lin LW. Growth of *Listeria monocytogenes* and *Pseudomonas fragi* on cooked pork in a modified atmosphere packaging/nisin combination system. J. Food Protect. 1994; 57(6):479–485.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

11. Fang TJ, Lin LW. Inactivation of *Listeria monocytogenes* on raw pork treated with modified atmosphere packaging and nisin. J. Food Drug Anal. 1994;2(3):189–200.

12. Farber JM, Cai Y, Ross WH. Predictive modeling of the growth of *Listeria monocytogenes* in CO_2 environments. Int. J. Food Microbiol. 1996; 32(1–2):133–144.

13. Farber JM. Microbiological aspects of modified-atmosphere packaging technology — a review. J. Food Protect. 1991; 54(1):58–70.

14. Farber JM, Daley E. Fate of *Listeria monocytogenes* on modified-atmosphere packaged turkey roll slices. J. Food Protect.1994; 57(12):1098–1100.

15. Fernandez PS, George SM, Sills CC, Peck MW. Predictive model of the effect of CO_2, pH, temperature and NaCl on the growth of *Listeria monocytogenes*. Int. J. Food Microbiol. 1997; 37(1):37–45.

16. Grant IR, Patterson MF. Effect of irradiation and modified atmosphere packaging on the microbiological safety of minced pork stored under temperature abuse conditions. Int. J. Food Sci. Technology. 1991; 26(5):521–533.

17. Hudson JA, Mott SJ, Penney N. Growth of *Listeria monocytogenes, Aeromonas hydrophila*, and *Yersinia enterocolitic*a on vacuum and saturated carbon dioxide controlled atmosphere-packaged sliced roast beef. J. Food Protect. 1994; 57(3):204–208.

18. Ingham SC, Escude JM, McCown P. Comparative growth rates of *Listeria monocytogenes* and *Pseudomonas fragi* on cooked chicken loaf stored under air and two modified atmospheres. J. Food Protect. 1990; 53(4):289–291.

19. Juven BJ, Barefoot SF, Pierson MD, McCaskill LH, Smith B. Growth and survival of *Listeria monocytogenes* in vacuum-packaged ground beef inoculated with *Lactobacillus alimentarius floracarn* l-2. J. Food Protect. 1998; 61(5):551–556.

20. Kim KT, Murano EA, Olson DG. Heating and storage conditions affect survival and recovery of *Listeria monocytogenes* in ground pork. J. Food Sci. 1994; 59(1):30–32, 59.

21. Kraemer KH, Baumgart J. Sliced frankfurter-type sausage. Inhibiting *Listeria monocytogenes* by means of a modified atmosphere. Fleischwirtschaft 1993; 73(11):1279–1280.

22. Laack RLJM van, Johnson JL, Palen CJNM van der, Smulders FJM, Snijders JMA. Survival of pathogenic bacteria on pork loins as influenced by hot processing and packaging. J. Food Protect. 1993; 56(10):847–851, 873.

23. Lee M, Sebranek JG, Olson DG, Dickson JS. Irradiation and packaging of fresh meat and poultry. J. Food Protect. 1996; 59(1):62–72.

24. Mano SB, Defernando GDG, Lopez-Galvez D, Selgas MD, Garcia ML, Cambero MI, Ordonez JA. Growth survival of natural flora and *Listeria monocytogenes* on refrigerated uncooked pork and turkey packaged under modified atmospheres. J. Food Safety. 1995; 15(4):305–319.

25. Manu Tawiah W, Myers DJ, Olson DG, Molins RA. Survival and growth of *Listeria monocytogenes* and *Yersinia enterocolitica* in pork chops packaged under modified gas atmospheres. J. Food Sci. 1993; 58(3):475–479.

26. Marshall DL, Andrews LS, Wells JH, Farr AJ. Influence of modified atmosphere packaging on the competitive growth of *Listeria monocytogenes* and *Pseudomonas fluorescens* on precooked chicken. Food Microbiol. 1992; 9(4):303–309.

27. Marshall DL, Wiese-Lehigh PL, Wells JH, Farr AJ. Comparative growth of *Listeria monocytogenes* and *Pseudomonas fluorescens* on precooked chicken nuggets stored under modified atmospheres. J. Food Protect. 1991; 54(11):841–843.

28. Michel ME, Keeton JT, Acuff GR. Pathogen survival in precooked beef products and determination of critical control points in processing. J. Food Protect. 1991; 54(10):767–772.

29. Miller MF, Carr MA, Schluter AR, Jones DK, Meade MK, Ramsey, CB. Distribution packaging method and storage time effects on the microbiological characteristics and incidence of the pathogens *Listeria monocytogenes* and *Salmonella* in pork. J. Food Qual. 1996; 19(5):413–422.

30. Phillips CA. Review: Modified atmosphere packaging and its effects on the microbiological quality and safety of produce. Int. J. Food Sci. Technol. 1996; 31(6):463–479.

31. Sheridan JJ, Doherty A, Allen P, McDowell DA, Blair IS, Harrington D. Growth/survival of psychrotrophic pathogens on meat packaged under modified atmospheres. Food Microbiol. 1995; 12(3):259–266.

32. Thayer DW, Boyd G. Irradiation and modified atmosphere packaging for the control of *Listeria monocytogenes* on turkey meat. J. Food Protect. 1999; 62(10):1136–1142.

33. Wimpfheimer L, Altman NS, Hotchkiss JH. Growth of *Listeria monocytogenes* Scott A, serotype 4 and competitive spoilage organisms in raw chicken packaged under modified atmospheres and in air. Int. J. Food Microbiol. 1990; 11(3/4):205–214.

34. Zeitoun AAM, Debevere JM. Inhibition, survival and growth of *Listeria monocytogenes* on poultry as influenced by buffered lactic acid treatment and modified atmosphere packaging. Int. J. Food Microbiol. 1991; 14(2):161–169.

USE OF HIGH PRESSURE TO CONTROL *LISTERIA* IN MEAT

High hydrostatic pressure causes widespread damage to cells with adverse effects on membranes, enzymes and other structures and molecules (*7*). *Listeria monocytogenes* is sensitive to high pressure treatments of 400–500 MPa , but like other Gram-positive organisms (such as *Staphylococcus aureus*) is one of the more resistant species of bacteria. Some strain variation in sensitivity to pressure is evident at lower temperatures (25°C) but largely disappears at 50°C (*2*). Bacterial spores are very barotolerant, requiring pressures as high as 1000 MPa to destroy them (*5,13*).

With current interest in minimally processed foods, high pressure treatment has become a more attractive technique because of its minimal effect on the characteristics of the final product. The effects of high pressure are instantaneously and uniformly transmitted throughout foods regardless of their geometry or size. Although high pressure destroys living cells, it does not degrade small molecules like vitamins and flavors and (under the conditions tested) has minimal effects on the sensory quality of meats (*3,8*). One disadvantage is the difficulty in completely sterilizing foods; pressure destruction curves usually demonstrate some tailing, and damaged but viable cells may recover and start growing during storage (*10,13,16*). Nevertheless, several pressure-treated foods are currently being marketed, including fruit juices and jams and raw squid.

According to experiments with *L. monocytogenes* and *L. innocua* in laboratory media, several factors affect the lethality of a given level of high pressure. Modest increases in temperature (from 25 to 50°C) decreased D values from 50.8 to 22.4 min at 137.9 MPa and from 14.3 to 1.3 min at 344.7 MPa (*1,2*). Therefore, a 7 log kill could be achieved by exposure to 345 MPa pressure for approximately 9 minutes at 50°C. Increased acidity also enhances the effect of pressure. At 45°C and 252 MPa for 30 min, an 8 log kill of *L. monocytogenes* occurred at pH 4.0 and only a 2 log kill at pH 6.0 (*14*). Addition of the bacteriocin pediocin AcH also increases the effectiveness of high pressure: an 8 log reduction in *L. monocytogenes* cells was achieved in only 5 min at 345 MPa in the presence of pediocin (*6*). Compounds similar to those in foods (bovine serum albumin, glucose, and olive oil) exert a protective effect on *L. monocytogenes* as indicated by larger D values (*11*).

Experiments with minced beef, chicken, and pork and with pork chops inoculated with *L. monocytogenes* or other bacteria have confirmed the increased lethality caused by a moderate rise in temperature (*4,8,10,12*) and the presence of bacteriocins (*15,17*). Generally, somewhat higher pressures (400–500 MPa) were required to achieve a useful kill rate in a reasonable length of time in meat as compared to laboratory media. Some representative D values are presented in the table below.

It should be emphasized that effectiveness of high pressure treatments depends on temperature, length of exposure, and pressure intensity as well as the strain of *Listeria* used and various ingredients in different foods.

D values (min) in meat treated with high pressure

Bacteria*	Pressure	Temp. (°C)	Meat	D value	Ref. no.
L. monocytogenes	414 MPa	25	ground pork	4.17	8
L. monocytogenes	414 MPa	50	ground pork	0.63	8
L. monocytogenes	400 MPa	4	pork chop	3.52	9
L. monocytogenes	414 MPa	25	pork chop	2.17	3
S. typhimurium	414 MPa	25	pork chop	1.48	3
L. monocytogenes	375 MPa	18	raw chicken	5.0	12
L. monocytogenes	375 MPa	18	cooked chicken	9.2	12
L. monocytogenes	375 MPa	18	raw minced beef	4.9	12
L. monocytogenes	375 MPa	18	cooked minced beef	9.4	12
L. innocua	330 MPa	20	ground beef	6.5	4

**L. = Listeria; S. = Salmonella*

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

REFERENCES

1. Alpas H, Kalchayanand N, Bozoglu F, Ray B. Interaction of pressure, time and temperature of pressurization on viability loss of *Listeria innocua*. World J. Microbiol. Biotechnol. 1998; 14(2):251–253.

2. Alpas H, Kalchayanand N, Bozoglu F, Sikes A, Dunne CP, Ray B. Variation in resistance to hydrostatic pressure among strains of food-borne pathogens. Appl. Environ. Microbiol. 1999; 65(9):4248–4251.

3. Ananth, V, Dickson, JS, Olson, DG, Murano EA. Shelf life extension, safety, and quality of fresh pork loin treated with high hydrostatic pressure. J. Food Protect. 1998; 61(12):1649–1656.

4. Carlez A, Rosec JP, Richard N, Cheftel JC. High pressure inactivation of *Citrobacter freundii*, *Pseudomonas fluorescens* and *Listeria innocua* in inoculated minced beef muscle. Lebensm. Wiss. Technol. 1993; 26(4):357–363.

5. Earnshaw RG, Appleyard J, Hurst RM. Understanding physical inactivation processes — combined preservation opportunities using heat, ultrasound and pressure. Int. J. Food Microbiol. 1995; 28(2):197–219.

6. Kalchayanand N, Sikes A, Dunne CP, Ray B. Interaction of hydrostatic pressure, time and temperature of pressurization and pediocin AcH on inactivation of foodborne bacteria. J. Food Protect. 1998; 61(4):425–431.

7. Mackey BM, Forestiere K, Isaacs NS, Stenning R, Brooker B. The effect of high hydrostatic pressure on *Salmonella thompson* and *Listeria monocytogenes* examined by electron microscopy. Lett. Appl. Microbiol. 1994; 19(6):429–432.

8. Murano EA, Murano PS, Brennan RE, Shenoy K, Moreira, R. G. Application of high hydrostatic pressure to eliminate *Listeria monocytogenes* from fresh pork sausage. J. Food Protect. 1999; 62(5):480–483.

9. Mussa DM, Ramaswamy HS, Smith JP. High-pressure destruction kinetics of *Listeria monocytogenes* on pork. J. Food Protect. 1999; 62(1):40–45.

10. Patterson MF, Kilpatrick DJ. The combined effect of high hydrostatic pressure and mild heat on inactivation of pathogens in milk and poultry. J. Food Protect. 1998; 61(4):432–436.

11. Simpson RK, Gilmour A. The effect of high hydrostatic pressure on *Listeria monocytogenes* in phosphate-buffered saline and model food systems. J. Appl. Microbiol. 1997; 83(2):181–188.

12. Simpson RK, Gilmour A.. The resistance of *Listeria monocytogenes* to high hydrostatic pressure in foods. Food Microbiol. 1997; 14(6):567–573.

13. Smelt JPPM. Recent advances in the microbiology of high pressure processing. Trends Food Sci. Technol. 1998; 9(4):152–158.

14. Stewart CM, Jewett FF, Dunne CP, Hoover DG. Effect of concurrent high hydrostatic pressure, acidity and heat on the injury and destruction of *Listeria monocytogenes*. J. Food Safety. 1997; 17(1):23–36.

15. ter Steeg PF, Hellemons JC, Kok AE. Synergistic actions of nisin, sublethal ultrahigh pressure, and reduced temperature on bacteria and yeast. Appl. Environ. Microbiol. 1999; 65(9):4148–4154.

16. Yuste J, Mor-Mur M, Capellas M, Pla R. *Listeria innocua* and aerobic mesophiles during chill storage of inoculated mechanically recovered poultry meat treated with high hydrostatic pressure. Meat Sci. 1999; 53(4):251–257.

17. Yuste J, Mor-Mur M, Capellas M, Guamis B, Pla R. Microbiological quality of mechanically recovered poultry meat treated with high hydrostatic pressure and nisin. Food Microbiol. 1998; 15(4):407–414.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

USE OF PULSED ELECTRIC FIELDS AND ELECTROLYZED OXIDIZING WATER TO CONTROL *LISTERIA*

Pulsed electric field (PEF) pasteurization is a non-thermal process which destroys contaminating bacteria by short bursts (< 1 sec) of high voltage. Exposure to PEF destabilizes cell membranes and with sufficient intensity and duration of treatment, membranes are irreversibly damaged, important cellular compounds leak out, and cells die (*2*,*6*). At lower PEF doses, these effects on cell membranes have been exploited by genetic engineers to induce hybridization of cells and introduction of DNA fragments into cells (*2*).

Bacterial spores, Gram-positive cells (including *L. monocytogenes*), and cells in stationary phase of growth are more resistant to the effects of PEF (*1*). For *L. monocytogenes* suspended in milk, a continuous flow PEF system resulted in a 3 log reduction in bacterial numbers at 25°C and a 4 log decrease at 50°C (*5*). A model of microbial survival after exposure to PEF has been developed (*3*).

As yet this new technology has been applied primarily to liquids such as juices, milk, yogurt, beaten eggs, sauces, and soups (*4*). A PEF system has also been used to destroy *E. coli* in a homogeneous semisolid medium (potato dextrose agar) (*8*). Pumpable food pastes such as vegetable or fruit purées and minced meat are also possible candidates for this type of pasteurization (*1*). Bacteria in dry powders (flour, spices), however, appear to be less susceptible to PEF compared to those in liquids (*2*). Further research is needed to determine the potential for use of PEF for the pasteurization of viscous and particulate foods.

Electrolyzed oxidizing water (EO water) is acidic water (≤ pH 2.7) collected from the anode during electrolysis of deionized water containing a low concentration of NaCl. Tests demonstrated that a 2 min exposure of *L. monocytogenes* to EO water at 35°C resulted in ≥ 7 log kill (*7*). This process is still in early developmental stages but may have future applications in food processing.

An excellent review of the methods of PEF and its prospects for use in food processing is presented by Barsotti and Cheftel (*1*).

REFERENCES

1. Barsotti L, Cheftel JC. Food processing by pulsed electric fields. II Biological aspects. Food Rev. Int. 1999; 15(2):181–213.

2. Jeyamkondan S, Jayas DS, Holley RA. Pulsed electric field processing of foods: a review. J. Food Protect. 1999; 62(9):1088–1096.

3. Peleg M. A model of microbial survival after exposure to pulsed electric fields. J. Sci. Food Agric. 1995; 67(1):93–99.

4. Qin BL, Pothakamury UR, Vega H, Martin O, Barbosa-Canovas GV, Swanson BG. Food pasteurization using high-intensity pulsed electric fields. Food Technol. 1995; 49(12):55–60.

5. Reina LD, Jin ZT, Zhang, QH, Yousef AE. Inactivation of *Listeria monocytogenes* in milk by pulsed electric field. J. Food Protect. 1998; 61(9):1203–1206.

6. Simpson RK, Whittington R, Earnshaw RG, Russell NJ. Pulsed high electric field causes 'all or nothing' membrane damage in *Listeria monocytogenes* and *Salmonella typhimurium*, but membrane H+-ATPase is not a primary target. Int. J. Food Microbiol. 1999; 48(1):1–10.

7. Venkitanarayanan KS, Ezeike GO, Hung YC, Doyle MP. Efficacy of electrolyzed oxidizing water for inactivating *Escherichia coli* O157:H7, *Salmonella enteritidis*, and *Listeria monocytogenes*. Appl. Environ. Microbiol. 1999; 65(9):4276–4279.

8. Zhang QH, Chang FJ, Barbosa-Canovas V, Swanson BG. Inactivation of microorganisms in a semisolid model food using high voltage pulsed electric fields. Lebensm. Wiss. Technol. 1994; 27:538–543.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

USE OF ULTRAVIOLET LIGHT TO CONTROL *LISTERIA* IN MEAT

Although the bactericidal effects of ultraviolet light have long been utilized to control microbial contamination in some medical and food industry areas, it is only recently that techniques using UV to reduce the microbial load on foods, such as the surfaces of meat, have been developed. Since UV light cannot penetrate into foods, only microbes on an exposed surface are susceptible to its effects. Bacteria on a smooth surface such as agar plates in laboratories or flat plate beef absorb more UV light than bacteria on a rough surface such as some cuts of beef, pork, or chicken skin (*3*,*5*,*6*). Therefore, the UV exposure required for effective killing of bacteria on meat will most likely exceed that required for killing cells on laboratory media.

Studies have shown that UV exposure does not have a deleterious effect on the color of meat nor does it cause oxidative rancidity (*3*,*5*). This is because UV light does not induce production of oxidizing free radicals. Rather, the toxicity of UV light is primarily due to the formation of thymine dimers which disrupt the structure and functioning of DNA in bacterial cells.

Experiments with *Listeria monocytogenes* demonstrated that cells in a moist environment were killed more easily than those in a dry film or crust (*7*). In addition, shorter wavelengths (254 nm) were more effective than longer wavelengths (365 nm) of UV light (*7*). In the presence of psoralen compounds (from parsley, limes, celery, etc.), longer wavelengths of UV can kill *L. monocytogenes* and other bacteria (*4*).

A recent innovation which greatly increases the peak power in the UV light source is the pulse power energization technique (PPET). PPET light sources operating at 1 pulse/second kill *L. monocytogenes* on an agar surface much faster than a continuous light source and can reduce cell populations by 6 logs in a 512 µs (*1*). PPET sources can be developed to operate at 100–1000 pulses/sec and these high energy sources may be practical for disinfecting meat surfaces.

A comparison of susceptibility of foodborne pathogens (grown on agar plates) revealed that *L. monocytogenes* was the most resistant to UV light (*2*): *L. monocytogenes* > *Staphylococcus aureus* $\geq$ *Salmonella enteritidis* > *E. coli* > *Bacillus cereus*.

REFERENCES

1. MacGregor SJ, Rowan NJ, McIlvaney L, Anderson JG, Fouracre RA, Farish O. Light inactivation of food-related pathogenic bacteria using a pulsed power source. Lett. Appl. Microbiol. 1998; 27(2):67–70.

2. Rowan NJ, MacGregor SJ, Anderson JG, Fouracre RA, McIlvaney L, Farish O. Pulsed-light inactivation of food-related microorganisms. Appl. Environ. Microbiol. 1999; 65(3):1312–1315.

3. Stermer RA, Lasater-Smith M, Brasington CF. Ultraviolet radiation – an effective bactericide for fresh meat. J. Food Protect. 1987; 50(2):108–111.

4. Ulate-Rodriguez J, Schafer HW, Zottola EA, Davidson PM. Inhibition of *Listeria monocytogenes*, *Escherichia coli* O157-H7, and *Micrococcus luteus* by linear furanocoumarins in culture media. J. Food Protect. 1997; 60(9):1046–1049.

5. Wallner-Pendleton EA, Sumner SS, Froning GW, Stetson LE. The use of ultraviolet radiation to reduce *Salmonella* and psychrotrophic bacterial contamination on poultry carcasses. Poultry Sci. 1994; 73:1327–1333.

6. Wong E, Linton RH, Gerrard DE. Reduction of *Escherichia coli* and *Salmonella senftenberg* on pork skin and pork muscle using ultraviolet light. Food Microbiol. 1998; 15:415–423.

7. Yousef AE, Marth EH. Inactivation of *Listeria monocytogenes* by ultraviolet energy. J. Food Sci. 1988; 53(2):571–573.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999

USE OF ULTRASOUND TO CONTROL *LISTERIA* IN MEAT

The lethal effects of ultrasound have been known since sonar was developed to detect submarines and nearby fish were killed. Ultrasound kills by disrupting cell membranes apparently as a result of the formation and subsequent implosion of small bubbles (cavitation). Heat and some chemicals may enhance the lethal effects of ultrasound.

Currently, ultrasound is used in food processing for emulsification, accelerating freezing and cleaning (*1*). Some recent investigations have focussed on the possible uses of ultrasound for the destruction of foodborne pathogens. Because viscous liquids and solids impede the propagation of ultrasound waves, this technique is potentially most useful for sterilization of liquids, such as milk and juices. At some future time, ultrasound, in combination with other preservation methods, may be useful in surface sterilization of other foods. Therefore, some recent research papers describing the effects of combinations of ultrasound and high pressure and/or heat on *L. monocytogenes* in liquids have been listed below (*2*,*3*,*4*).

REFERENCES

1. Earnshaw RG, Appleyard J, Hurst RM. Understanding physical inactivation processes — combined preservation opportunities using heat, ultrasound and pressure. Int. J. Food Microbiol. 1995; 28(2):197–219.

2. Pagan R, Manas P, Alvarez I, Condon S. Resistance of *Listeria monocytogenes* to ultrasonic waves under pressure at sublethal (manosonication) and lethal (manothermosonication) temperatures. Food Microbiol. 1999; 16(2):139–148.

3. Pagan R, Manas P, Palop A, Sala FJ. Resistance of heat-shocked cells of *Listeria monocytogenes* to mano-sonication and mano-thermo-sonication. Lett. Appl. Microbiol. 1999; 28(1):71–75.

4. Pagan R, Manas P, Raso J, Condon S. Bacterial resistance to ultrasonic waves under pressure at nonlethal (manosonication) and lethal (manothermosonication) temperatures. Appl. Environ. Microbiol. 1999; 65(1):297–300.

Prepared by M. Ellin Doyle, Ph.D.
Food Research Institute, University of Wisconsin–Madison

November 1999