Epidemiology of Human Listeriosis

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INTRODUCTION

Listeria monocytogenes is a gram-positive bacillus found frequently in the environment that causes stillbirths and meningococcal infanticides in a wide range of animals (53). Human disease due to L. monocytogenes usually occurs in the setting of pregnancy, immunosuppression, or extremes of age. Epidemiologic investigation of several outbreaks of listeriosis during the 1980s (12, 41, 93, 145) demonstrated that epidemic listeriosis is a foodborne disease. Recent study suggests that a substantial portion of sporadic listeriosis cases may also be caused by foodborne organisms (23, 147). Laboratory advances in detection and subtyping of the organism have recently improved efforts to study human listeriosis.

The successful use of immunosuppressive medications in the treatment of malignancy and the management of organ transplantation, as well as the more recent epidemic of acquired immunodeficiency syndrome, have led to an expansion of the immunosuppressed population at increased risk of listeriosis. Attention has therefore been directed to listeriosis, both as a clinical entity of increasing importance and as a substantial problem for the food industry.

In this article, we discuss recent developments in microbiologic detection and subtyping of L. monocytogenes and review current information on epidemic and sporadic disease, specifically detailing each of the major outbreaks caused by foodborne organisms in the past decade. We also discuss recent efforts to determine the magnitude of disease due to L. monocytogenes in the United States and to identify risk factors for sporadic listeriosis. The proceedings of two recent symposia on listeriosis provide detailed reviews of virulence factors and immunologic aspects of infection (1, 2).

MICROBIOLOGY OF L. MONOCYTOGENES

Identification and Biochemical Characteristics

Listeria spp. are gram-positive rods 0.4 to 0.5 μm in diameter and 0.5 to 2 μm in length (151). Members of the genus Listeria are aerobic and facultatively anaerobic, do not produce spores, and demonstrate characteristic motility when cultured at 20 to 25°C. Colonies are bluish gray by normal illumination, but a blue-green sheen is visible by oblique light. Listeria spp. can grow between pH 6 and pH 9 and in temperatures ranging from 1 to 45°C. Optimum

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growth occurs between 30 and 37°C. *Listeria* spp. are catalase positive, oxidase negative, methyl red positive, and Voges-Proskauer positive.

*Listeria monocytogenes* is beta-hemolytic on blood agar and forms a narrow zone of hemolysis around colonies. In contrast, *L. ivanovi* forms double or triple hemolytic zones when grown on sheep or horse blood agar, and other *Listeria* species are nonhemolytic. The CAMP test is useful in differentiating hemolytic species of *Listeria* and in separating *L. innocua* from *L. monocytogenes*, since these two species give similar reactions to biochemical tests. The CAMP test detects synergistic reactions of hemolysins of *Listeria* spp. with the beta toxin of *Staphylococcus aureus* and with an exofactor of *Rhodococcus equi*. *Listeria monocytogenes* gives a positive CAMP reaction on sheep blood agar with *Staphylococcus aureus* but not *Rhodococcus equi*, while *L. ivanovi* produces a positive CAMP reaction with *R. equi* but not *S. aureus*. *L. innocua* gives a negative CAMP reaction with *S. aureus* and *R. equi*. All *Listeria monocytogenes* strains produce acid from L-rhamnose and α-methyl-D-mannoside but not from D-xylose or D-mannitol. Table 1 summarizes features distinguishing *L. monocytogenes* from other *Listeria* species.

**Isolation of *Listeria monocytogenes***

*L. monocytogenes* is readily isolated from clinical specimens obtained from normally sterile sites such as cerebrospinal fluid, blood, and amniotic fluid. *L. monocytogenes* grows on a wide variety of nonselective plating media (e.g., blood agar, chocolate agar, tryptic soy agar, and brain heart infusion agar). When *L. monocytogenes* is present in high numbers (10^6 CFU/ml), direct plating on these media yields virtually pure cultures (159).

The isolation of *Listeria monocytogenes* from foods and clinical specimens that are not usually sterile requires selective enrichment before specimens are plated on selective isolation agars. Ever since Gray et al. (54) discovered the psychrotrophic characteristics of *L. monocytogenes*, cold enrichment (4°C) in nonselective broth medium has been used to enhance its isolation. However, cold enrichment for 1 to several weeks may be required for isolation of the organism from foods and clinical specimens (59, 73). Further, other psychrotrophic bacteria present in foods may overgrow *Listeria* spp. during cold enrichment (71). Because of these disadvantages of cold enrichment, other selective enrichment procedures have been developed for the isolation of *Listeria* spp. from foods.

Chemicals used to render liquid enrichment media selective for *L. monocytogenes* include acriflavine, glycyne hydroxide, lithium chloride, nalidixic acid, nitrofurazone, potassium tellurite, and potassium thiocyanate (83). Among the most frequently used selective chemical enrichment media are (i) Food and Drug Administration (FDA) broth, an enrichment broth prepared from tryptic soy broth, acriflavine, and nalidixic acid (95, 97); (ii) U.S. Department of Agriculture-Food Safety and Inspection Service (USDA-FSIS) broth, containing esculin, acriflavin, and nalidixic acid; developed by Dominguez Rodriguez et al. (31), modified by Donnelly and Baigent (32), and used by the USDA-FSIS for the isolation of *L. monocytogenes* from meats and poultry products (109); (iii) nutrient broth containing 3.75% potassium thiocyanate and nalidixic acid, developed by Watkins and Sleath (167) and used by Hayes et al. (59) to isolate *L. monocytogenes* from epidemic-associated raw milk; and (iv) L-PALCAM broth, a medium developed in The Netherlands (165) and used widely in Europe. Foods are enriched in these media for 24 to 48 h and plated on *Listeria*-selective agars. Hayes et al. (59) used the potassium thiocyanate-nalidixic acid-containing medium for the secondary enrichment of cold-enriched cultures.

McBride and Girard (108) developed the first selective plating agar for *L. monocytogenes* Their formulation was blood-containing medium composed of phenylethanol agar base, lithium chloride, and glycine. Lovett (95) used a modification of the McBride agar (without blood) to isolate *L. monocytogenes* from foods at the FDA laboratories. Researchers at USDA-FSIS improved the selectivity of the McBride medium by increasing the concentration of lithium chloride 10-fold, substituting glycin anhydride for glycine, and incorporating the antibiotic moxalactam (91). This formulation, commonly known as LPM agar, is highly selective and is useful for the isolation of *L. monocytogenes* from highly contaminated specimens. The FDA amended its *Listeria* isolation procedure (164) by including LPM agar for plating enrichment cultures. Neither the modified McBride’s agar nor LPM agar contain any chemicals that would allow for the differential presumptive identification of *Listeria* colonies by colony morphology and color. Growth on these media is examined under oblique 45° illumination to detect the typical light blue colonies formed by *Listeria* species. The PALCAM medium developed by van-Netten et al. (165) and the Oxford agar developed by Curtis et al. (27) attempt to overcome this disadvantage by incorporating differential agents in the agar medium. On PALCAM agar, *Listeria* colonies appear grey-green, are approximately 2 mm in diameter, and have black, sunken centers. They also have a black halo against a cherry red background (165). On Oxford agar, *L. monocytogenes* colonies appear black, are 1 mm (24 h) to 3 mm (48 h) in diameter, and are surrounded by black halos (27). PALCAM and Oxford agars are preferred by

**TABLE 1. Characteristics of *Listeria* spp.**

<table>
<thead>
<tr>
<th>Species</th>
<th>Hemolysis</th>
<th>Acid production from:</th>
<th>Nitrate reduction</th>
<th>Result of CAMP test with:</th>
</tr>
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<tr>
<td></td>
<td></td>
<td>d-Glucose</td>
<td>d-Xylose</td>
<td>d-Mannitol</td>
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<tr>
<td><em>L. monocytogenes</em></td>
<td>+</td>
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<td><em>L. innocua</em></td>
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<tr>
<td><em>L. ivanovi</em></td>
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<tr>
<td><em>L. seeligeri</em></td>
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<tr>
<td><em>L. welshimeri</em></td>
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<td><em>L. grayi</em></td>
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<td><em>L. murrayi</em></td>
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* Adapted from reference 168. +, ≥90% of strains were positive; −, <90% of strains were negative; V, 11 to 89% of strains were positive.
those who have difficulty observing the blue color of *Listeria* spp. on LPM and McBride agars under oblique illumination. Oxford agar was useful for the isolation of *L. monocytogenes* from milk and dairy products (124).

Several investigators have compared the various selective enrichment and plating methods. Many comparative studies suffer from poor design and very low positivity rates, limiting the statistical evaluation of the results. Pini and Gilbert (132) compared the cold enrichment and FDA procedures for the isolation of *L. monocytogenes* from soft cheeses and raw chickens. They found that the FDA procedure was more productive for the isolation of *L. monocytogenes* from chickens, while the methods were essentially equivalent for isolation of *L. monocytogenes* from cheeses. In contrast, Doyle and Schoeni (34) found cold enrichment to be superior to the FDA procedure for the isolation of *L. monocytogenes* from soft cheeses. Parish and Higgins (128) found the FDA procedures for the isolation of *L. monocytogenes* from 402 soft cheeses. Lammerding and Doyle (52, 154, 155) suggested the need for additional methods. In addition, the method provides information on the degree of genetic relatedness between strains. In determining whether a relationship between a clinical and an environmental isolate exists, it is important to consider the background distribution of subtypes in the environment. Such background information on distribution of enzyme types was recently obtained for a well-characterized sample of *L. monocytogenes* isolates acquired during population-based surveillance in the United States (11, 49). Similar information has been collected for a large but less well defined sample of clinical and environmental isolates from several countries (131); however, 41 of the 114 human isolates characterized in the report were recovered from patients with a common-source listeriosis outbreak in Switzerland. Both studies suggest that electrophoretic enzyme typing permits *L. monocytogenes* to be divided into a large number of distinct subtypes. Because multilocus enzyme electrophoresis permits typing of all strains and because substantial diversity exists among *Listeria* strains, the method is extremely useful in epidemiologic investigations that attempt to compare clinical and environmental strains.

Perez-Diaz et al. (129) examined 32 *Listeria* strains from species *L. monocytogenes*, *L. murrayi*, and *L. grayi* for the presence of plasmids and found a 38.5-MDa cryptic plasmid in 4 strains of *L. monocytogenes*, 2 strains of *L. grayi*, and 1 strain of *L. murrayi*. Restriction analysis of the plasmids indicated that all were a single molecular species. Analysis of plasmids may not provide adequate discrimination of *Listeria* species and therefore may not be suitable for subtyping *L. monocytogenes*.

Restriction fragment length polymorphism (RFLP) analysis was applied by Nocera et al. (123) to 28 strains of *L. monocytogenes* serotype 4b. They obtained 10 different RFLP profiles with EcoRI restriction enzyme. However, four different RFLP profiles were observed for 19 strains involved in a single outbreak that were a single clone according to multilocus electrophoretic enzyme typing. Wesley et al. (175) used *Hinf* restriction enzyme to examine *L. monocytogenes* serotype 4b strains from the four North American outbreaks for RFLP of genomic DNA. They obtained distinctly different patterns for strains from different outbreaks but identical profiles for strains from the same outbreak. While RFLP analysis appears to be useful for the characterization of *L. monocytogenes*, the high degree of discrimination offered by this procedure may not always be epidemiologically relevant.

One of the problems of RFLP analysis is that a large number of bands (100 to 1,000) are obtained by restriction of genomic DNA with restriction enzymes with four- or six-base recognition sequences. This makes it difficult to apply RFLP analysis to a large set of strains to determine the degree of interrelatedness. Attempts have been made to
simplify RFLP analysis by performing hybridization experiments on the restriction fragments of chromosomal DNA with probes that recognize relatively stable chromosomal regions. Saunders et al. (141) used two cloned probes selected from a bacteriophage lambda library of L. monocytogenes to probe NcoI digests of L. monocytogenes DNA from 64 strains. They found 19 different patterns and observed similar profiles in epidemiologically related strains.

Ribosomal DNA fingerprinting (ribotyping) has also been used to subtype L. monocytogenes. Ribotyping involves probing restriction fragments of chromosomal DNA with 16S and 23S rRNA of Escherichia coli. This method has two distinct advantages. Because ribosomal genes are highly conserved across phyla, one universal probe can be used for the characterization of several organisms (56). Also, because bacteria usually contain multiple rRNA operons (seven in E. coli), an adequate number of bands (5 to 15) of different molecular sizes are obtained by this procedure. Swaminathan et al. (158) characterized 89 strains of L. monocytogenes by ribotyping. Sixteen different ribotypes were observed. In general, strains with a strong epidemiologic association were found to be the same ribotype. Further, ribotyping divided the strains into two distinct subgroups (separating serotypes 1/2a, 1/2c, and 3a from serotypes 1/2b and 4b) in the same manner observed with multilocus enzyme electrophoresis. Ribotyping in conjunction with enzyme electrophoresis provided laboratory support for a suspected association between an isolate of L. monocytogenes from raw milk and a patient with listeriosis who drank the milk (166).

Although several methods of subtyping L. monocytogenes have been investigated, most subtyping methods are labor intensive and do not easily permit rapid screening of a large number of isolates. Questions concerning the reproducibility and sensitivity of various typing methods continue to be investigated.

Rapid Confirmation of L. monocytogenes

Colonies that give a Listeria-like appearance on primary plating media are subjected to biochemical tests for identification of L. monocytogenes. Confirmation of presumptively positive colonies as Listeria species or as L. monocytogenes by conventional methods requires 2 to 7 days. This is problematic when a rapid answer is required, such as in the quality control of semiperishable food products which have a limited shelf life.

Several procedures have been proposed for the rapid confirmation of Listeria species or of L. monocytogenes. Fraser and Sperber (43) modified the USDA secondary enrichment broth by supplementing it with lithium chloride and ferric ammonium citrate. Listeria species hydrolyze esculin in the USDA secondary enrichment broth, producing 6,7-dihydroxycoumarin. The ferric ions react with this compound to produce a black precipitate. Hydrolysis of the esculin by enterococci in the sample is prevented by the addition of lithium chloride to the medium. Kempton (77) proposed the use of a sodium chloride-esculin hydrolysis test, motility test, and hemolysis test to rapidly identify Listeria species. The use of a petri plate with four quadrants (quad plate) has been proposed as an aid for rapid identification of Listeria species (140). The quad plate allows the performance of five tests: oxidase, catalase, CAMP test, rhamnose, and xylose (159). Another procedure for rapid confirmation, the API 20 STREP system, was successfully used to differentiate L. monocytogenes and L. seeligeri from L. ivanovii and nonhemolytic Listeria species in 4 h (101).

DNA probes derived from the listeriolysin O gene sequence (24, 115), the gene coding for a putative invasion factor of L. monocytogenes (28, 29, 40, 80), and the gene for a delayed-type hypersensitivity factor (125) of L. monocytogenes have been used for the rapid confirmation of L. monocytogenes colonies isolated from food. Mengaud et al. (115) and Chenever et al. (24) found a 651-bp internal fragment of the listeriolysin gene to be specific for L. monocytogenes under stringent hybridization conditions. Datta et al. (28) reported that pRF106, a 321-bp internal fragment of a gene coding for a 60-kDa secreted polypeptide in L. monocytogenes, was specific for L. monocytogenes under moderately stringent hybridization conditions; however, Kim et al. (80) reported that the same probe hybridized to two of five strains of L. seeligeri even at stringent hybridization conditions. An oligonucleotide probe derived from the nucleotide sequence of pRF106 reacted with L. monocytogenes and one strain of L. seeligeri (29). Interestingly, an oligonucleotide probe that differed in only one nucleotide from the probe used by Datta et al. (29) did not react with the same L. seeligeri at stringent hybridization conditions (79). A DNA probe derived from the nucleotide sequence of a putative delayed-type hypersensitivity factor of L. monocytogenes reacted with 175 of 186 strains of L. monocytogenes tested but also with L. ivanovii under stringent hybridization conditions. An L. monocytogenes-specific acidimium ester-labeled DNA probe was used in a homogeneous protection assay for the confirmation of L. monocytogenes colonies in less than 2 h (126). This method may be very useful for the rapid confirmation of food isolates.

Rapid Detection of Listeria Species and L. monocytogenes

Monoclonal antibody-based immunoassays and nucleic acid hybridization assays have been developed for the rapid detection of Listeria spp. in foods and in clinical specimens. Foods generally contain low concentrations of Listeria spp. in relation to the total microbial load. Therefore, the analysis of foods by these methods is typically done after the sample has been cultured in one or more selective enrichment broths.

Several monoclonal antibody-based tests are available for the rapid detection of Listeria species. Monoclonal antibodies to cell surface antigens are only genus specific; these have been used to develop a dot-enzyme immunoassay, a microplate enzyme immunoassay, and a direct immunofluorescence test for the rapid detection of Listeria species (19, 37, 107, 113, 114). Although none of the monoclonal antibodies to cell surface antigens are specific exclusively to L. monocytogenes, monoclonal antibodies against the beta-hemolysin (listeriolysin O) of L. monocytogenes are specific and could potentially be used in the rapid detection and confirmation of L. monocytogenes (61).

DNA probe assays have also been developed for the rapid detection of Listeria species but are not yet available for the rapid and specific detection of L. monocytogenes from enrichment broth cultures of foods. A commercially available DNA probe assay (Gene-Track Systems, Framingham, Mass.) uses a fluorescein-labeled DNA probe targeted to a Listeria species-specific rRNA sequence for rapid detection of Listeria species in foods (82, 84).

A two-step nested polymerase chain reaction has been developed to detect L. monocytogenes DNA in paraffin-
embedded tissues (81). The diagnostic fragment is a 165-bp internal fragment of the listeriolysin gene. The method, if validated in further evaluations, may offer a means of detecting L. monocytogenes DNA retrospectively in preserved clinical specimens and could potentially be modified for food applications.

**L. MONOCYTOGENES IN THE ENVIRONMENT**

*L. monocytogenes* has been identified throughout the environment. The organism has been isolated from soil (171), water, and decaying vegetation (170, 172). Weis detected *L. monocytogenes* in 21% of 779 plant and soil samples; serotypes 1/2b and 4b were found most frequently (170). The ability of *L. monocytogenes* to survive in the environment has been demonstrated quantitatively by Watkins and Sleath (167), who isolated *L. monocytogenes* from samples of sewage, river water, and sewage sludge by using cold enrichment methods. They found that quantitative counts of *L. monocytogenes* from sewage sludge sprayed on agricultural land remained unchanged for at least 8 weeks. The potential implications of using fecal material as agricultural fertilizer subsequently became evident when this practice was suspected to have contributed to a large outbreak of human listeriosis in Nova Scotia (145).

**L. MONOCYTOGENES IN ANIMALS**

Beginning with Murray and colleague’s description in 1926 of disease in rabbits caused by the bacterium now known as *L. monocytogenes* (120), listeriosis has generally been thought of as a veterinary disease. In mammals, *L. monocytogenes* causes abortions and “circling disease” (meningoencephalitis), and epizootics of listeriosis were observed in herds of cattle and sheep long before outbreaks of listeriosis were recognized in humans. In addition, healthy animals could be gastrointestinal carriers of *L. monocytogenes*.

*L. monocytogenes* has been isolated from cattle, pigs, sheep, chickens, turkeys, ducks, and a variety of other species (149). Systematic culture of stool from cows with *Listeria*-related abortions, from healthy cows in herds with listeriosis, and from cows in unaffected herds revealed differences in the carriage of *L. monocytogenes* (13). The highest rates were found in stool cultures of animals who had aborted because of listeriosis (24% [53 of 219 samples]), while healthy cows from affected and unaffected herds had lower rates of carriage (affected herds, 6.7% [41 of 622]; unaffected herds, 1.7% [2 of 120]) (13). There may be a relationship between the feeding of poor-quality silage and the onset of listeriosis in domesticated ruminants.

Because clinical illness and asymptomatic carriage of *L. monocytogenes* were well documented in animals, listeriosis was considered a zoonosis; human disease was thought to be due to illness occurring in animals, and the animal host was considered the primary reservoir for the organism (13). Consistent with this view, conjunctivitis was reported in poultry workers who handled infected chickens (38), and cutaneous lesions have been reported among veterinarians and cattle ranchers who delivered infected and aborted calves (20, 127). In most reported cases of human illness, however, there is no history of direct contact with animals. Gray and Killinger stressed this discrepancy in their observation that most human listeriosis occurred in urban residents, while only rare cases occurred among residents of rural areas where domestic animals were widely affected by listeriosis (53).

The idea that human listeriosis could be the result of indirect contact with infected animals focused attention on the possibility of transmission by foodborne organisms. Case reports had suggested that human listeriosis occurred after consumption of contaminated foods such as unpasteurized milk from cows known to be infected with the organism (133). However, *Listeria* species may be isolated frequently from the environment or from foods without a direct relation to human illness, and initial typing systems were not sufficiently precise to suggest that an environmental strain was related to a clinical isolate. Investigation of large outbreaks of human listeriosis finally provided epidemiologic and laboratory support to confirm the suspicion that listeriosis was a foodborne disease (12, 41, 93, 145).

**L. MONOCYTOGENES IN HUMANS**

**Carriage**

As early as 1926, Murray et al. (120) suggested that the intestinal tract might be the portal of entry for organisms causing *Listeria* infections. While most studies of nasopharyngeal cultures from healthy individuals failed to detect *Listeria* spp. (105, 157), several investigators have found *L. monocytogenes* in fecal specimens from healthy people (13, 55, 70, 72, 73, 88), a fact consistent with the suggestion that the gastrointestinal tract is the human reservoir of the organism.

Bojsen-Moller used cold enrichment to study fecal carriage in a number of population groups (13). He found *Listeria* in stool cultures from healthy slaughterhouse workers (4.8% [55 of 1,147 workers]), hospitalized adult patients (1.2% [12 of 1,034 patients]), patients with diarrhea (1% [6 of 595 patients]), and household contacts of listeriosis patients (26% [9 of 34 household contacts]). Because up to eight specimens were collected from individual household contacts of patients, the frequency of *Listeria* isolation in this group is not directly comparable to the results from other populations. Five of 14 households had at least one member with *L. monocytogenes* in stool. However, in only two of the households was the family member carrying the same serotype as the patient. Cold enrichment was used in processing all the cultures, but transport of specimens to a central laboratory was delayed in the nonhospitalized patients and antibiotic use prior to culture was more frequent in the hospitalized patients, which complicated comparison between groups.

Other studies from various pregnant and nonpregnant populations have identified wide-ranging estimates for stool carriage of *L. monocytogenes* among healthy adults. Kampmacher and van Noorle Jansen (72) found that 11.9% of office personnel sampled had fecal *Listeria* carriage, and slaughterhouse workers had a rate of 13.3%. Gregorio and Eveland (55) found *L. monocytogenes* in 1.75% of stool samples from 400 patients hospitalized with nonlisteric conditions. Differences in culture methods as well as in dietary and host factors may account for differences in the prevalence of carriage. Higher rates of *Listeria* carriage have been reported when serial specimens from individual subjects were cultured, but comparing such estimates of cumulative prevalence with estimates of point prevalence is problematic.
Invasive Disease

Pathogenesis. *L. monocytogenes* can be found frequently in the environment, and carriage studies suggest that human exposure to *L. monocytogenes* is not uncommon, yet invasive listeriosis occurs rarely. Factors that may influence whether invasive disease will occur include the virulence of the infecting organism, the susceptibility of the host, and the size of the inoculum.

Transmission of *L. monocytogenes* by food first requires penetration of the organism through the intestine (10). Intracellular multiplication can occur in various types of cells. Mackaness first showed that *L. monocytogenes* could grow inside nonimmune phagocytes (102), and more-recent experimental work suggests that *Listeria* spp. can replicate in Peyer's patches (100). Gaillard et al. (44, 45) showed in an in vitro model using human enterocytes like cells that *L. monocytogenes* multiplies intracellularly after internalization into vacuoles. This directed phagocytosis was observed for *L. monocytogenes* and *L. ivanovi*, the pathogenic species of *Listeria*, but not for nonpathogenic species of *Listeria* (45). Quantitative electron microscopic study suggests that *L. monocytogenes* moves from phagocytic vacuoles to the cytoplasm, where replication is improved by more-favorable growth conditions. The ability to disrupt vacuolar membranes, therefore, is considered important in the virulence of *L. monocytogenes*.

Transposon mutagenesis experiments suggest that a hemolysin, listeriolysin O, permits pathogenic *Listeria* species to escape the phagosome (46, 74). Nonhemolytic variants were avirulent in a mouse model, and virulence was restored in a revertant strain which gained the ability to produce hemolysin after spontaneous loss of the transposon (46, 74). Listeriolysin O was not involved in bacterial internalization, since nonhemolytic strains penetrated cells in vitro at the same rate as hemolytic strains (45). Progress in elucidating the role of hemolysin in the virulence of *L. monocytogenes* has included purification of listeriolysin O (50) and measurement of cytolytic activity under various experimental conditions. Cytolytic activity was greatest at pH 5.5 and undetectable at pH 7.0, suggesting that lytic activity would be mainly expressed in the phagolysosome rather than in extracellular fluids (50).

Sword demonstrated that growth of *L. monocytogenes* in mice and host susceptibility to experimental infection were correlated with the availability of iron to the organism (160). In contrast, iron deprivation stimulates hemolysin secretion. Iron deprivation within the phagosome favors production of listeriolysin O, leading to disruption of intracellular membranes. Iron availability and neutral pH in the cytoplasm therefore favor bacterial replication after escape of the bacteria from the vacuole. Although listeriolysin O appears to be important in virulence, levels of hemolysin do not directly correlate with virulence in experimental infection in mice (76), and discrepancies between experimental results in murine and human cell lines still need to be clarified (75).

Host susceptibility to *L. monocytogenes* depends primarily on cell-mediated immunity, and most listeriosis occurs in persons with impaired cell-mediated immunity due to disease processes, medications, or pregnancy. *L. monocytogenes* was used by Mackaness in early studies that delineated the nature of cell-mediated immunity (102). Protection in mice against *L. monocytogenes* is mediated through *Listeria*-sensitized T cells, which activate macrophages. Mackaness showed that macrophage activation depended on specific lymphocytes (103) and that antilymphocyte globulin suppressed immunity to *Listeria* spp. (104). The main function of *Listeria*-sensitized T cells appears to be attracting, focusing, and activating macrophages at infective foci (116). T cells activate macrophages by producing lymphokines including gamma interferon, which has been shown to increase the listericidal activity of dexamethasone-treated monocytes (142). Prostaglandins may mediate suppression of cellular immunity to *L. monocytogenes* (130, 163). Macrophage-derived thromboxane A2, via vasoconstriction, decreases the numbers of bacteria that can leave the site of infection; treatment with the cyclooxygenase inhibitor indomethacin was shown to increase the susceptibility of mice to *Listeria* infection (163). Cytotoxic suppressor T cells appear to have a more important role in eradicating *Listeria* spp. than do helper cells (116).

Pregnancy-associated depression in cell-mediated immunity may be due to alterations in hormonal and serum factors as well as to a decrease in the ratio of T helper to suppressor cells during pregnancy (169). In addition, the local placental suppression of cell-mediated immunity necessary to prevent maternal rejection of the placenta may contribute to susceptibility of the fetus to infection with listeriosis (138).

In addition to depressed cell-mediated immunity, deficiencies in immunoglobulin M and complement activity associated with the neonatal state may contribute to the propensity of infants to develop listeriosis (14). Local gastrointestinal factors may play an additional role in disease in adults (62, 148).

Although the infectious dose of listeriosis in humans is not known, host susceptibility probably influences the size of the inoculum that can cause infection. The lethal dose of *L. monocytogenes* is reduced substantially in steroid-treated mice (117). Outbreaks of disease caused by foodborne listeriae in which the majority of nonpregnant adults were not immunodeficient (145) might conceivably involve a higher infectious dose of *Listeria* spp. than outbreaks in which illness occurred only in immunosuppressed persons (46, 62, 148). Oral-feeding experiments with Sprague-Dawley rats suggest that infection is dose dependent and that gastric acidity is protective; cimetidine-treated rats were susceptible to lower inocula of *L. monocytogenes* than animals with normal gastric acid (144).

Invasive disease in nonpregnant adults. In 1967, Louria et al. first described an association between listeriosis and malignant disease (94), and more recent reviews suggest that most invasive listeriosis occurs in persons who are immunosuppressed or elderly (111, 122, 147). Use of immunosuppressive medications for the management of malignancies and in organ transplantation has increased the immunosuppressed population and brought increased attention to listeriosis in the medical literature (122). With the epidemic of acquired immunodeficiency syndrome (AIDS) and the resulting rapid expansion of the population at substantial risk for listeriosis, this disease may be more frequently diagnosed.

In their review of adult listeriosis cases reported in the literature from 1968 to 1978, Nieman and Lorber (122) found that most cases of meningitis or bacteremia due to *L. monocytogenes* occurred in persons with malignancies (27% [40 of 148]) or receiving immunosuppressive treatments for nonmalignant conditions (31% [46 of 148]). Other conditions such as alcoholism, diabetes, and cirrhosis accounted for lower proportions of cases. However, in 30% of meningitis patients and 11% of bacteremic patients, no predisposing condition was recognized.

In a more-recent study of sporadic listeriosis cases occur-
ring in 1986 to 1987 in a geographically diverse population in the United States, 88% of nonperinatal cases occurred in persons with one or more underlying diseases (147). The most common conditions were cancer (23%), diabetes (20%), renal disease (18%), and heart disease (17%); several patients had more than one underlying condition.

Additional underlying illnesses that have been reported in association with listeriosis include sarcoidosis, chronic otitis, collagen-vascular disease, idiopathic thrombocytopenic purpura, asthma, ulcerative colitis, and aplastic anemia (122).

Several investigators have reported listeriosis among persons with AIDS or human immunodeficiency virus infection and have commented on the surprisingly "infrequent" occurrence of listeriosis among this population with substantial deficiency of cell-mediated immunity (58, 67, 69, 106, 136, 137). Although it is not one of the more-common infections experienced by human immunodeficiency virus-infected patients, listeriosis does occur approximately 300 times more frequently in persons with AIDS than in the general population (49, 173).

Nonpregnant adults with listeriosis most often present with meningitis, meningoencephalitis, or sepsis. Additional syndromes include abscesses of the brain (30) and spinal cord (119), endocarditis (7, 47), endophthalmitis (6), osteomyelitis (63), and septic arthritis (17). Fever, ataxia, seizures, depressed consciousness, and altered mental status are frequent presenting symptoms of central nervous system listeriosis. Spinal fluid examination may show pleocytosis with predominantly polymorphonuclear leukocytes; Gram stain may show gram-positive bacilli but is more often unrevealing; the protein level is elevated; and the glucose level usually is within normal limits. However, many other patterns have been observed. Diagnosis is made by culture of L. monocytogenes from spinal fluid, blood, or some other usually sterile site.

**Listeriosis during pregnancy.** Listeriosis may develop at any time during pregnancy, although most infections are detected in the third trimester. Infections occurring earlier in pregnancy may not be recognized if cultures are not obtained, and failure to culture the products of conception and placenta may eventually lead to improved understanding of patterns that have been observed. Diagnosis is made by culture of L. monocytogenes from fetal fluids, blood, or some other usually sterile site.

**Noninvasive Disease: Mild Syndrome due to Listeriosis.**

Because listeriosis presents with invasive syndromes such as meningitis and stillbirth, the association of the illness with a foodborne organism has not always been apparent. Listeriosis differs from other foodborne diseases in which noninvasive syndromes have been well characterized. One may speculate that a milder gastrointestinal illness might occur when persons without underlying immunosuppression eat food contaminated by L. monocytogenes. If this were the case, the numbers of cases of illness caused by Listeria spp. would be much greater than previously estimated. However, investigating mild illness associated with listeriosis has been difficult. In Bojesen-Møller's study of carriage in the household contacts of patients with listeriosis, establishing an association between symptoms and Listeria excretion was not possible (13). Three of the persons with stool carriage of L. monocytogenes may have had diarrhea at the time the sample was obtained. However, information on gastrointestinal symptoms of controls was not obtained.

The question of noninvasive illness was recently investi-
gated among persons who had attended a catered party following which two guests delivered infants with listeriosis caused by the same strain of *L. monocytogenes* (139). Many of the partygoers experienced mild gastrointestinal symptoms. However, stool cultures were not obtained until several weeks after the party, and the outbreak strain of *L. monocytogenes* was isolated from only one additional guest. *Listeria* carriage and gastrointestinal symptoms could therefore not be correlated.

**EPIDEMIOLOGIC PATTERNS OF DISEASE**

**Epidemic Disease**

**Community outbreaks: foodborne disease.** A large outbreak in the Maritime Provinces of Canada provided the first evidence for transmission of listeriosis by foodborne organisms (145). The outbreak was recognized when perinatal listeriosis occurred in 1.3% of births at the maternity hospital in Halifax, Nova Scotia. Between March and September 1981, there were 7 adult and 34 perinatal cases of listeriosis, including 5 spontaneous abortions and 4 stillbirths. The case fatality rate among liveborn infants was 27%. In contrast to most patients with sporadic disease, nonpregnant adult patients with listeriosis in this outbreak had no evidence of an underlying immunosuppressive condition.

A case-control study conducted during the investigation suggested that patients were more likely than controls to have eaten coleslaw prior to their illness ($P = 0.02$). Coleslaw from the refrigerator of one patient grew *L. monocytogenes* serotype 4b, the same serotype as the epidemic strain. The coleslaw was prepared in the area, and two unopened packages of the coleslaw from the manufacturer also grew the organism. The manufacturer had purchased cabbage from a farmer who also raised sheep. Two of the farmer’s sheep had died of listeriosis the year before the outbreak. The farmer used composted and raw sheep manure to fertilize his cabbage fields, which may have led to contamination of the crop. Cabbage was stored over the winter and spring in a large shed; growth of *L. monocytogenes* was probably enhanced by prolonged cold storage. The outbreak investigation documented that listeriosis is a foodborne disease and suggested potential problems with consumption of uncooked vegetables.

Another outbreak of listeriosis may also have involved raw vegetables. In September and October 1979, 20 patients were hospitalized in Boston with listeriosis due to serotype 4b; only 9 cases had been detected in the preceding 26 months (62). Fifteen of 20 patients had hospital-acquired infection. Patients were more likely than controls to have eaten tuna fish, chicken salad, and cheese, although several brands were used to prepare each of these foods. However, each food was served with a raw-vegetable garnish, which may have been the vehicle of listeriosis, although these garnishes were not known to be from a common supplier.

Though no single common food exposure was implicated in this outbreak, the investigation suggested that conditions in the gastrointestinal tract may alter the risk of an individual's developing listeriosis. Use of antacids or histamine-blocking drugs was more frequent among patients than controls. As has been observed for salmonellosis, it is possible that the lack of gastric acidity increases the chance that *L. monocytogenes* will survive passage through the stomach. In this outbreak, patients were also more likely than controls to have undergone gastrointestinal procedures prior to culture of *L. monocytogenes*. Enteritis associated with *Listeria* infection might have caused gastrointestinal symptoms which prompted gastrointestinal procedures, or underlying gastrointestinal lesions could have increased the risk that intraluminal exposure to *L. monocytogenes* would cause invasive disease.

A second large outbreak in Massachusetts occurred from 30 June to 30 August 1983 (41). Most infections occurred in nonpregnant adults, all of whom had immunosuppressive conditions. In addition, there were seven perinatal cases. The overall case fatality rate was 29%. Serotype 4b accounted for 32 of the 40 isolates that could be tested. Patients were more likely than controls matched for neighborhood or underlying disease to have drunk a specific brand of pasteurized whole or 2% fat milk. Additional evidence that pasteurized milk caused the outbreak was the presence of a dose-response effect, a protective effect from skim milk consumption, implication of the same product in a separate study done in a different state, and implication of a specific phage type (2425A) of *L. monocytogenes* in patients who had drunk the milk. Listeriosis was known to have occurred in dairy cows that provided milk to the implicated manufacturer. Though *L. monocytogenes* (not of the epidemic type) was isolated from raw milk from these farms, inspections of the plant were postpasteurization. The outbreak raised concern that pasteurization may be inadequate to kill *Listeria* spp. and led to extensive research concerning the heat resistance of the bacterium (see below). Postpasteurization contamination is one likely explanation for this large outbreak.

The largest epidemic of listeriosis in North America occurred in 1985 in Los Angeles, Calif. (93). The majority of infections occurred in pregnant women or their offspring. Perinatal cases included 87 infants with early-onset infections or stillborn and 6 late-onset cases. The case fatality rate was 63% for early neonatal or fetal infections and 37% for nonneonatal infections.

Eighty-seven percent (81 of 93) of pregnancy-associated cases occurred in Hispanic women, and a case-control study implicated a specific brand of Mexican-style soft cheese as the vehicle of disease. *L. monocytogenes* serogroup 4b, of the same phage type as the epidemic strain, was isolated from unopened packages of this brand of cheese. In June 1985, the product was recalled and the factory was closed. Contamination of the cheese with unpasteurized milk probably led to the outbreak.

As calculated from food consumption histories from four patients who ate the implicated cheese only once, the incubation period for listeriosis was a median of 31 days (range, 11 to 70 days). The interval between eating a contaminated food and onset of symptoms appears to be much longer for listeriosis than for other foodborne diseases, which typically cause symptoms from hours to a few days after exposure to contaminated food. Investigation of sporadic listeriosis is therefore quite complicated, since relevant food exposures may be those which occurred several weeks prior to the onset of disease.

Because the majority of cases in the Los Angeles outbreak occurred in a single ethnic group primarily cared for at a single medical facility, the outbreak was recognized quickly and the implicated food was recalled during the investigation. Much apparently sporadic listeriosis may be the result of contaminated food sources, but temporal clustering of cases may not be recognized if the vehicle is widely distributed or if patients present to different medical facilities. Distinguishing sporadic disease from common-source out-
breaks can be facilitated with serotyping and subtyping of strains. In addition, enhanced surveillance for listeriosis can improve early recognition of outbreaks. In 1986, the Council of State and Territorial Epidemiologists recommended that listeriosis be made a reportable disease.

Soft cheese was responsible for another outbreak of listeriosis, centered in Switzerland and recognized in 1987 (12). The outbreak was traced to a regionally produced soft smear cheese which was distributed to several countries in Europe. Identification of \( L.\ monocyto genes \) in the implicated product led to an international food recall. The \( Listeria \) strain that caused this outbreak was the same electrophoretic enzyme type as the strain responsible for the outbreak associated with Mexican soft cheese (131). Whether certain strains of \( Listeria \) preferentially grow in soft cheeses or other foods or are of enhanced virulence is not known.

An unusual outbreak of listeriosis occurred in Philadelphia, Pa., in 1987 (148). Most (32 of 36) patients were nonpregnant adults, the majority of whom had immunosuppressive conditions or were elderly. Only 3 of the 36 patients had no immunosuppressive risk factor for listeriosis. Although the clustering of cases represented a threefold increase over the background incidence of listeriosis (20 cases per million population versus 7 cases per million), the investigation suggested that this increase was not the result of a single contaminated food source. Patients were more likely than controls to have eaten ice cream or salami prior to disease onset, but no single brand of either food was implicated. Of a total of 22 available isolates from patients in the outbreak revealed that multiple strains were involved. Eleven different enzyme types were identified, including several different enzyme types from patients who had eaten salami and ice cream. The investigation demonstrated the important role that subtyping may play in clarifying epidemiologic observations.

The investigation revealed that patients were more likely than controls to suffer gastrointestinal symptoms prior to diagnosis of listeriosis. One-third of patients had vomiting and diarrhea. In addition, more of the family members of patients than of controls were ill in the month before onset of listeriosis in the patient. Although these symptoms in the patients and their family members could have been due to an enteral phase of listeriosis infection, the investigators speculated that the presence of a coinfesting organism such as an enteric virus might have converted asymptomatic gastrointestinal carriage of \( L.\ monocyto genes \) to invasive listeriosis. An enteric virus circulating in a community might increase the risk of individuals with sporadic exposure to \( L.\ monocytogenes \) developing disease and consequently might cause an outbreak of disease associated with multiple strains of \( L.\ monocytogenes \).

Nosocomial outbreaks. In addition to the large community outbreaks attributed to contaminated food sources, several nosocomial clusters of neonatal listeriosis have been reported (21, 39, 42, 89, 98, 121, 153). Most of the reports describe the delivery of an infant with early-onset infection followed by the diagnosis of late-onset listeriosis in one or more infants born subsequently (21, 42, 66, 68, 89, 121, 153). In most reports, person-to-person transmission caused by inadequate handwashing and breaks in barrier nursing technique was considered to have caused late-onset infections (21, 66, 68, 98). In some reports, the affected infants had common exposure to resuscitation equipment (121, 153), and in some hospitals, inadequate cleaning of a shared rectal thermometer may have caused two cases of necrotizing enterocolitis due to \( L.\ monocytogenes \), although transmission from person to person could not be ruled out (89). Nosocomial clusters have been small, and microbiologic efforts have failed to document any common source that was suggested epidemiologically.

Investigation of a larger nosocomial cluster of listeriosis in 1989 in Costa Rica provided the opportunity to evaluate risk factors for late-onset disease in addition to the vehicle and modes of transmission responsible for the outbreak (146). More than 3% of infants born during the outbreak period developed listeriosis. The outbreak investigation suggested that transmission occurred when newborns were bathed with mineral oil from a common container, from which the outbreak strain of \( L.\ monocytogenes \) was cultured. The outbreak occurred following the delivery of an infant with early-onset disease born to a mother with amniotitis. The oil presumably became contaminated during the delivery of the source patient. Although all newborns were bathed with contaminated oil during the subsequent period, those born by cesarian section were more likely to develop disease. Transmission may have occurred when infants aspirated contaminated oil applied on the face or when oil came in contact with mucous membrane surfaces. The respiratory portal of entry was supported by respiratory presenting symptoms in several infants and the finding of lipid-laden macrophages, consistent with oil aspiration, in lung tissue from a patient who died of the disease. General anesthesia used in cesarian deliveries may have increased the infants’ risk of aspirating the oil they were exposed to shortly after birth. The outbreak underscored the ability of \( L.\ monocytogenes \) to persist in the hospital environment once introduced, confirming the important role of multiuse materials in transmission of infectious diseases in the delivery room.

**Sporadic Listeriosis**

**Surveillance.** The numbers of cases of disease due to \( L.\ monocytogenes \) have been difficult to determine. Voluntary reporting to state health departments and analysis of hospital discharge data have been used to attempt to estimate the incidence of disease in the United States (25, 118). However, such methods have low sensitivity and are likely to underestimate the true rate of disease.

To obtain a more precise estimate of the incidence of listeriosis in the United States, the Centers for Disease Control (CDC) performed active surveillance for listeriosis in 1986 in five states and Los Angeles County (total population, 34 million) (49). The surveillance project detected an annual incidence of 0.7 cases per 100,000 population. On the basis of this study, CDC estimates that 1,700 cases of listeriosis occur annually in the United States and result in 450 adult deaths and 100 fetal and postnatal deaths. The overall rate of perinatal listeriosis was 12.7 cases per 100,000 live births, but Los Angeles County had a rate of perinatal listeriosis significantly higher than that of other areas (24.3/100,000 births versus 7.8/100,000 births). Race-specific rates of perinatal listeriosis for both blacks and whites were higher in Los Angeles County than elsewhere.

These differences could reflect true geographic differences in incidence, or they might be the result of differences in obstetric practices that could lead to enhanced diagnosis of perinatal listeriosis in Los Angeles. In 1985, the year preceding the CDC surveillance project, a well-publicized outbreak of listeriosis had occurred in Los Angeles (93). Increased awareness of the disease and more aggressive attempts at diagnosis by physicians in the area may have
improved detection of listeriosis, resulting in the higher rates of disease observed in that area.

The surveillance project did not detect geographic differences in the rates of nonperinatal listeriosis (49). The incidence of listeriosis increased with age; 84% of the patients with nonperinatal listeriosis were over 50 years old, and 41% were 70 years or older. In contrast to studies conducted in tertiary care centers, the surveillance project found that bacteremia was a more common presentation of nonperinatal listeriosis than was meningitis. This difference could reflect a true change in the presentation of illness from earlier studies, or it could be a manifestation of differences between the general population and persons referred to tertiary care centers.

**Dietary risks for sporadic disease.** In addition to determining the magnitude of listeriosis in the United States, the CDC active surveillance project was used to evaluate dietary risk factors for sporadic disease (147). Patients detected by active surveillance were enrolled in a case-control study that compared dietary histories of persons who will listeriosis and those of controls matched for age, underlying illness or pregnancy, and health care provider (an indirect measure of geographic location and socioeconomic status). Persons who had eaten undercooked chicken or uncooked hot dogs were more likely to develop listeriosis. Though this epidemiologic study did not include laboratory investigation of the actual foods eaten by patients with listeriosis, the results had substantial plausibility. In various microbiologic surveys of retail foods, *L. monocytogenes* was cultured from 15 to 80% of poultry samples and 30% of ready-to-eat meat products (179).

Subsequent investigation of sporadic listeriosis has provided microbiologic support for the initial epidemiologic study. In one instance, listeriosis was associated with consumption of microwaved turkey franks (23). The same electrophoretic enzyme type of *L. monocytogenes* was isolated from a patient with listeriosis, from a package of turkey franks in the patient’s refrigerator, and from unopened franks from a local store. Inspection of the factory resulted in detection of the same strain of *L. monocytogenes* in cultures of environmental surfaces and in final container packages ready for sale (174).

Investigation of sporadic listeriosis has also suggested that cross-contamination of food products occurs in the home (23). The patient’s refrigerator which contained the implicated turkey franks also contained several other opened food items from which the same enzyme type of *L. monocytogenes* was cultured. As is the case with salmonellosis, careful food handling may be as important as food production monitoring for the successful prevention of foodborne listeriosis.

**Possible other sources of sporadic disease.** Transmission by foodborne organisms may account for the largest portion of sporadic disease, but other modes of transmission may play some role in occasional cases. Although *Listeria* spp. have been isolated from urethral exudates (149), there is no evidence that sexual transmission contributes to perinatal listeriosis infection. Because *L. monocytogenes* can survive refrigeration and because bacteremia may be asymptomatic, transmission of listeriosis through blood transfusion theoretically could occur. Such transmission has been documented for *Yersinia enterocolitica* (161), but no such occurrence has been described for *L. monocytogenes*.

**DIAGNOSIS, TREATMENT, AND PREVENTION**

Listeriosis is diagnosed when *L. monocytogenes* is cultured from blood, spinal fluid, or some other normally sterile site. Isolation of *Listeria* spp. from nonsterile sites such as placenta and amniotic fluid in association with clinical symptoms may suggest perinatal listeriosis. Culture of *L. monocytogenes* from stool is not helpful in diagnosis, since gastrointestinal carriage of the organism may occur without clinical disease. Serologic tests do not contribute to diagnosis of listeriosis, because antibody response may not be specific (64) and because patients with culture-proven listeriosis have had undetectable levels of antibody to *L. monocytogenes* (145).

*L. monocytogenes* is susceptible to a number of antibiotics in vitro, including penicillin G, ampicillin, erythromycin, trimethoprim-sulfamethoxazole, chloramphenicol, rifampin, tetracyclines, and aminoglycosides. Although controlled clinical trials have not been performed, ampicillin or penicillin is usually recommended for invasive listeriosis. Ampicillin may be superior to penicillin (90). Addition of an aminoglycoside to a beta-lactam antibiotic produces synergy in vitro (176), and this combination therapy is considered the treatment of choice. Laboratory study suggests that chloramphenicol and rifampin may each antagonize the bactericidal effect of penicillins (177). Case reports describe the effectiveness of treating patients who are allergic to penicillin with trimethoprim and sulfamethoxazole (57, 143, 156); this therapy is bactericidal, and adequate drug levels are found in serum and cerebrospinal fluid (178). Although cephalosporins may be bacteriostatic against *L. monocytogenes*, treatment failures have been observed (162), and treatment with cephalosporins is not recommended. Duration of therapy for listeriosis has not been standardized. Reasonable guidelines range from 2 weeks of therapy for uncomplicated sepsis or meningitis to 4 to 6 weeks of therapy for endocarditis or nonnecrotizing disease in the immunocompromised host (48).

Recommendations to prevent listeriosis are still evolving. Persons at increased risk, such as pregnant women and immunosuppressed adults, should be advised to avoid eating unpasteurized milk products and raw or partially cooked meats (18). Consumption of raw eggs may be a risk factor for listeriosis (as well as salmonellosis) in some populations (147), and persons at risk should avoid these products. In addition, foods should be prepared without cross-contamination between raw and cooked foods, and vegetables should be washed carefully. Foods prepared in a microwave oven may be unevenly heated, and sufficient standing time after cooking may be important to permit even distribution of heat by conduction without overcooking (99). Internal temperatures alone may not be adequate to ensure the safety of all microwaved foods, and new cooking guidelines may need to be established to minimize the risk of infection associated with eating microwaved foods (92).

**ISSUES FOR THE FOOD INDUSTRY**

Unusual growth and survival properties of *L. monocytogenes* contribute to the complexity of producing *Listeria*-free foods. The ability of the organism to survive refrigeration and freezing has important implications. A low initial inoculum in a food at the time of manufacturing can translate into a substantial dose of listeriae for the consumer, depending on the shelf life and handling of a particular product. As new methods of food preparation are introduced to the
public (e.g., cook-chill methods [51, 78]), the potential for transmission of Listeria spp. needs to be considered. On the basis of epidemiologic and laboratory implications of processed meats as a cause for listeriosis (23, 147) and concern regarding growth of the organism in processed foods before use by the consumer, the USDA in 1989 instituted a zero tolerance policy for L. monocytogenes in ready-to-eat products.

Early reports suggested that Listeria spp. were relatively heat resistant (8). Because L. monocytogenes can be isolated from approximately 5% of unpasteurized milk samples (97, 179), the adequacy of pasteurization in eliminating the organism from milk is of more than academic interest. The issue became important when investigation of an outbreak of listeriosis in 1983 implicated pasteurized whole and 2% milk as the source of infection (41). Procedures at the milk-processing plant in that outbreak were consistent with regulatory guidelines for pasteurization, raising questions about the adequacy of pasteurization itself. Several studies have subsequently shown that L. monocytogenes is inactivated by standard pasteurization (9, 16, 22, 33, 35), although investigators have continued to study the effects of growth temperature and anaerobic conditions on the survival of L. monocytogenes (85, 97). Postpasteurization contamination and deviation from time and temperature guidelines for pasteurization may explain the presence of L. monocytogenes in pasteurized products. Efforts to prevent listeriosis require the enforcement of regulatory guidelines for pasteurization procedures and the elimination of sources of postpasteurization contamination.

Species of Listeria other than L. monocytogenes can be isolated frequently from foods and food plants, but such organisms do not hold the same pathogenic potential as L. monocytogenes. Therefore, the refinement of molecular methods for the rapid detection of L. monocytogenes that are specific at the species level will have important applications in the food industry.

The frequency with which L. monocytogenes can be found during environmental surveys has raised doubts that food manufacturers can effectively eliminate L. monocytogenes from the workplace. However, investigation of a turkey-processing plant associated with a case of human listeriosis yielded encouraging results (174). Listeria contamination of the plant was not widespread; the strain associated with disease was restricted to the peeler used to process turkey franks. Additional industrial studies may improve the ability to identify areas where Listeria clean-up will be most effective.

SUMMARY

During the 1980s, investigation of several large epidemics of listeriosis confirmed that transmission of L. monocytogenes in food causes human disease. Progress in laboratory detection and subtyping of the organism has enhanced our ability to compare human and environmental isolates of L. monocytogenes. Transmission by foodborne organisms is now recognized as causing both epidemic and sporadic listeriosis. Continued study of dietary risk factors associated with listeriosis is needed in order to develop dietary recommendations for the expanding population at increased risk of disease. Current research application of new molecular methods to the study of L. monocytogenes may improve the ability to diagnose pregnancy-associated disease and permit the rapid detection and control of L. monocytogenes in the food supply.

REFERENCES


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