176 *Listeria monocytogenes* Infections

Elizabeth L. Hohmann, Daniel A. Portnoy

Listeria monocytogenes is a food-borne pathogen that can cause serious infections, particularly in pregnant women and immunocompromised individuals. A ubiquitous saprophytic environmental bacterium, *L. monocytogenes* is also a facultative intracellular pathogen with a broad host range. Humans are probably accidental hosts for this microorganism. *L. monocytogenes* is of interest not only to clinicians but also to basic scientists as a model intracellular pathogen that is used to study basic mechanisms of microbial pathogenesis and host immunity.

MICROBIOLOGY

L. monocytogenes is a facultatively anaerobic, nonsporulating, gram-positive rod that grows over a broad temperature range, including refrigeration temperatures. This organism is motile during growth at low temperatures but much less so at 37°C. The vast majority of cases of human listerial disease can be traced to serotypes 1/2a, 1/2b, and 4. *L. monocytogenes* is weakly β-hemolytic on blood agar, and (as detailed below) its β-hemolysin is an essential determinant of its pathogenicity.

PATHOGENESIS

Infections with *L. monocytogenes* follow ingestion of contaminated food that contains the bacteria at high concentrations. The conversion from environmental saprophyte to pathogen involves the coordinate regulation of bacterial determinants of pathogenesis that mediate entry into cells, intracellular growth, and cell-to-cell spread. Many of the organism's pathogenic strategies can be examined experimentally in tissue culture models of infection (Fig. 176-1). Like other enteric pathogens, *L. monocytogenes* induces its own internalization by cells that are not normally phagocytic. Its entry into cells is mediated by host surface proteins classified as internalins. Internalin-mediated entry is important in the crossing of intestinal, blood-brain, and feto-placental barriers, although how *L. monocytogenes* traffics from the intestine to the brain or fetus is only beginning to be investigated. In a pregnant guinea pig model of infection, *L. monocytogenes* was shown to traffic from maternal organs to the placenta; surprisingly, however, it also trafficked from the placenta back to maternal organs. These data are consistent with a model in which miscarriage can be viewed as a host defense strategy to eliminate a nidus of infection.

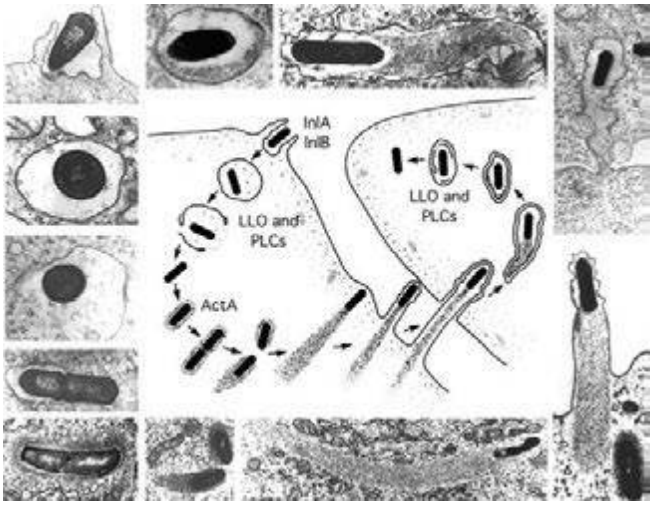

FIGURE 176-1 Stages in the intracellular life cycle of *Listeria monocytogenes*. The central diagram depicts cell entry, escape from a vacuole, actin nucleation, actin-based motility, and cell-to-cell spread. Surrounding the diagram are representative electron micrographs from which it was derived. ActA, surface protein mediating nucleation of host actin filaments to propel bacteria intra- and intercellularly; LLO, listeriolysin O; PLCs, phospholipases C; Inl, internalin. See text for further details. *(Adapted with permission from LG Tilney, DA Portnoy: J Cell Biol 109:1597, 1989. © Rockefeller University Press.)*

An essential determinant of the pathogenesis of *L. monocytogenes* is its β-hemolysin, listeriolysin O (LLO). LLO is a pore-forming, cholesterol-dependent cytolysin. (Related cytolysins include streptolysin O, pneumolysin, and perfringolysin O, all of which are produced by extracellular pathogens.) LLO is largely responsible for mediating the rupture of the phagosomal membrane that forms after phagocytosis of *L. monocytogenes*. LLO probably acts by insertion into an acidifying phagosome, which prevents the vesicle's maturation. In addition, LLO acts as a translocation pore for one or both of the *L. monocytogenes* phospholipases that also contribute to vacuolar lysis. LLO synthesis and activity are controlled at multiple levels to ensure that its lytic activity is limited to acidic vacuoles and does not affect the cytosol. Mutations in LLO that influence its synthesis, cytosolic half-life, or pH optimum cause premature toxicity to infected cells. There is an inverse relationship between toxicity and virulence—i.e., the more cytotoxic the strain, the less virulent it is in animals. This relationship may seem paradoxical, but, as an intracellular pathogen, *L. monocytogenes* benefits from leaving its host cell unharmed.

Shortly after exposure to the mammalian-cell cytosol, *L. monocytogenes* expresses a surface protein, ActA, that mediates the nucleation of host actin filaments to propel the bacteria intra- and intercellularly. ActA mimics host proteins of the Wiskott-Aldrich syndrome protein (WASP) family by promoting the actin nucleation properties of the Arp2/3 complex. Thus, *L. monocytogenes* can enter the cytosol of almost any eukaryotic cell or cell extract and can exploit a conserved and essential actin-based motility system. Other pathogens as diverse as certain *Shigella, Mycobacterium, Rickettsia*, and *Burkholderia* species use a related pathogenic strategy that allows cell-to-cell spread without exposure to the extracellular milieu.

IMMUNE RESPONSE

The innate and acquired immune responses to *L. monocytogenes* have been studied extensively in mice. Shortly after IV injection, most bacteria are found in Kupffer cells in the liver, with some organisms in splenic dendritic cells and macrophages. Listeriae that survive the bactericidal activity of initially infected macrophages grow in the cytosol and spread from cell to cell. *L. monocytogenes* triggers three innate immune pathways: a MyD88-dependent pathway leading to inflammatory cytokines, a STING/IRF3 pathway leading to a type I interferon response; and low-level inflammasome activation. Neutrophils are crucial to host defense during the first 24 h of infection, whereas an influx of activated macrophages from the bone marrow is critical subsequently. Mice that survive sublethal infection clear the organisms within a week, with consequent sterile immunity. Studies with knockout mice have been instrumental in dissecting the roles played by chemokines and cytokines during infection. For example, interferon γ, tumor necrosis factor, and CCR2 are essential in controlling infection. While innate immunity is sufficient to control infection, the acquired immune response is required for sterile immunity. Immunity is cell mediated; antibody plays no measurable role. The critical effector cells are cytotoxic (CD8+) T cells that recognize and lyse infected cells; the resulting extracellular bacteria are killed by circulating activated phagocytes.

A hallmark of the *L. monocytogenes* model is that killed vaccines do not provide protective immunity. The explanation for this fundamental observation is multifactorial, involving the generation of appropriate cytokines and the compartmentalization of bacterial proteins for antigen processing and presentation. Because the organism has the capacity to induce a robust cell-mediated immune response, attenuated strains have been engineered to express foreign antigens and are undergoing clinical studies as therapeutic vaccines for cancer.

EPIDEMIOLOGY

L. monocytogenes usually enters the body via the gastrointestinal tract in foods. Listeriosis is most often sporadic, although outbreaks