Listeria monocytogenes

Listeria monocytogenes is a bacterium that causes listeriosis, a disease that can have severe consequences for particular groups of the population. It can cause miscarriages in pregnant women and be fatal in immunocompromised individuals and the elderly. In healthy people, listeriosis generally only causes a mild form of illness. *L. monocytogenes* can be found throughout the environment. It has been isolated from domestic and wild animals, birds, soil, vegetation, fodder, water and from floors, drains and wet areas of food processing factories.

Description of the organism

L. monocytogenes is a Gram-positive, non-spore forming rod-shaped bacterium. It belongs to the genus *Listeria* along with *L. ivanovii, L. innocua, L. welshimeri, L. selligeri* and *L. grayi* (Rocourt and Buchrieser 2007). Of these species, only two are considered pathogens: *L. monocytogenes* which infects humans and animals, and *L. ivanovii* which infects ruminants (although there have been rare reports of *L. ivanovii* being isolated from infected humans) (Guillet et al. 2010). There are thirteen known serotypes of *L. monocytogenes*: 1/2a, 1/2b, 1/2c, 3a, 3b, 3c, 4a, 4ab, 4b, 4c, 4d, 4e and 7. The serotypes most often associated with human illness are 1/2a, 1/2b and 4b (FDA 2012).

Growth and survival characteristics

The growth and survival of *L. monocytogenes* is influenced by a variety of factors. In food these include temperature, pH, water activity, salt and the presence of preservatives (refer to Table 1).

The temperature range for growth of *L. monocytogenes* is between -1.5 and 45°C, with the optimal temperature being 30–37°C. Freezing can lead to a reduction in *L. monocytogenes* numbers (Lado and Yousef 2007). As *L. monocytogenes* can grow at temperatures as low as 0°C, it has the potential to grow, albeit slowly, in food during refrigerated storage.

Multiple factors influence the heat resistance of *L. monocytogenes*, including the characteristics of the food, such as salt content, water activiy and acidity. A higher fat content is more protective of *L. monocytogenes.* For example the D-value at 57.2°C for high fat beef (30.5%) and low fat beef (2%) was 5.8 and 2.6 minutes respectively; and for milk the D-value at 60°C in whole milk and skim milk was 1.5-2.1 and 0.95-1.05 minutes respectively. In vegetables, the D-value at 56°C ranged from 0.8 minutes for onions to 5.2 minutes for peas (Doyle et al 2001).

L. monocytogenes will grow in a broad pH range of 4.0–9.6 (Lado and Yousef 2007). Although growth at pH <4.0 has not been documented, *L. monocytogenes* appears to be relatively tolerant to acidic conditions. *L. monocytogenes* becomes more sensitive to acidic conditions at higher temperatures (Lado and Yousef 2007).

Like most bacterial species, *L. monocytogenes* grows optimally at a water activity (a_w) of 0.97. However, *L. monocytogenes* also has the ability to grow at a a_w of 0.90 (Lado and Yousef 2007). Johnson et al. (1988) demonstrated that *L. monocytogenes* can survive for extended periods of time at a a_w value of 0.81. *L. monocytogenes* is reasonably tolerant to salt and has been reported to grow in 13–14% sodium chloride (Farber et al. 1992). Survival in the presence of salt is influenced by the storage temperature. Studies have indicated that in concentrated salt solutions, the survival rate of *L. monocytogenes* is higher when the temperature is lower (Lado and Yousef 2007).

L. monocytogenes can grow under both aerobic and anaerobic conditions, although it grows better in an anaerobic environment (Sutherland et al. 2003; Lado and Yousef 2007).

The effect of preservatives on the growth of *L. monocytogenes* is influenced by the combined effects of temperature, pH, salt content and water activity. For example, sorbates and parabens are more effective at preventing growth of *L. monocytogenes* at lower storage temperatures and pH. Also, adding sodium chloride or lowering the temperature enhances the ability of lactate to prevent *L. monocytogenes* growth. At decreased temperatures (such as refrigeration storage) sodium diacetate, sodium propionate and sodium benzoate are more effective at preventing growth of *L. monocytogenes* (Lado and Yousef 2007).

Table 1: Limits for growth of *L. monocytogenes* when other conditions are near optimum (Lado and Yousef 2007)

	Minimum	Optimum	Maximum
Temperature (°C)	-1.5	30–37	45
pH	4.0	6.0–8.0	9.6
Water activity	0.90	0.97	–

Symptoms of disease

There are two main forms of illness associated with *L. monocytogenes* infection. Non-invasive listeriosis is the mild form of disease, while invasive listeriosis is the severe form of disease and can be fatal (FDA 2012). The likelihood that invasive listeriosis will develop depends upon a number of factors, including host susceptibility, the number of organisms consumed and the virulence of the particular strain (WHO/FAO 2004).

Symptoms of non-invasive listeriosis can include fever, diarrhoea, muscle aches, nausea, vomiting, drowsiness and fatigue. The incubation period is usually 1 day (range 6 hours to 10 days) (Painter and Slutsker 2007; FDA 2012). Non-invasive listeriosis is also known as listerial gastroenteritis or febrile listeriosis.

Invasive listeriosis is characterised by the presence of *L. monocytogenes* in the blood, in the fluid of the central nervous system (leading to bacterial meningitis) or infection of the uterus of pregnant women. The latter may result in spontaneous abortion or stillbirth (20% of cases) or neonatal infection (63% of cases). Influenza-like symptoms, fever and gastrointestinal symptoms often occur in pregnant women with invasive listeriosis. In non-pregnant adults, invasive listeriosis presents in the form of bacterial meningitis with a fatality rate of 30%. Symptoms including fever, malaise, ataxia, seizures and altered mental status (Painter and Slutsker 2007). The incubation period before onset of invasive listeriosis ranges from 3 days to 3 months (FDA 2012).

Virulence and infectivity

When *L. monocytogenes* is ingested, it may survive the stomach environment and enter the intestine where it penetrates the intestinal epithelial cells. The organism is then taken up by macrophages and non-phagocytic cells. The *L. monocytogenes* surface protein internalin is required for this uptake by non-phagocytic cells, as it binds to the receptors on the host cells to instigate adhesion and internalization. The bacterium is initially located in a vacuole after uptake by a macrophage or non-phagocytic cell. *L. monocytogenes* secrete listeriolysin O protein, which breaks down the vacuole wall and enables the bacteria to escape into the cytoplasm. Any bacteria remaining in the vacuole are destroyed by the host cell. Once located in the cytoplasm of the host cell, *L. monocytogenes* is able to replicate. *L. monocytogenes* is transported around the body by the blood, with most *L. monocytogenes* being inactivated when it reaches the spleen or liver. *L. monocytogenes* is able to utilise the actin molecules of the host to propel the bacteria into neighbouring host cells. In the case of invasive listeriosis, this ability to spread between host cells enables *L. monocytogenes* to cross the blood-brain and placental barriers (Montville and Matthews 2005; Kuhn and Goebel 2007; Bonazzi et al. 2009).

Mode of transmission

The most common transmission route of *L. monocytogenes* to humans is via the consumption of contaminated food. However, *L. monocytogenes* can be transmitted directly from mother to child (vertical transmission), from contact with animals and through hospital acquired infections (Bell and Kyriakides 2005).

Healthy individuals can be asymptomatic carriers of *L. monocytogenes*, with 0.6–3.4% of healthy people with unknown exposure to *Listeria* being found to shed *L. monocytogenes* in their faeces. However, outbreak investigations have shown that listeriosis patients do not always shed the organism in their faeces (FDA/USDA/CDC 2003; Painter and Slutsker 2007). Therefore the role of healthy carriers in the transmission of *L. monocytogenes* is unclear.

Incidence of illness and outbreak data

Listeriosis is a notifiable disease in all Australian states and territories. The incidence of listeriosis notified in Australia in 2012 was 0.4 cases per 100,000 population (93 cases). This is a slight increase from the previous 5 year mean of 0.3 cases per 100,000 population per year (ranging from 0.2–0.4 cases per 100,000 population per year) (NNDSS 2013). In Australia the fatality rate in 2010 was 21%, which was an increase from the 14% fatality rate of the previous year (OzFoodNet 2010; OzFoodNet 2012).

The notification rate for listeriosis in New Zealand in 2011 was 0.6 cases per 100,000 population (26 cases). This was an increase from the 2010 rate of 0.5 cases per 100,000 population. The fatality rate in New Zealand in 2011 was 3.8% (Lim et al. 2012).

In the United States (US) the notification rate for listeriosis in 2010 was 0.27 cases per 100,000 population. This was similar to the 2009 rate of 0.28 cases per 100,000 population (CDC 2012). In the European Union (EU) there were 0.32 confirmed cases of listeriosis per 100,000 population in 2011 (ranging from 0.04–0.88 cases per 100,000 population between countries). This was a 7.8% decrease in the number of cases from 2010. The reported fatality rate in the EU in 2011 was 12.7% (EFSA 2013).

Invasive *L. monocytogenes* infections can be life threatening, with average fatality rates being 20–30% among hospitalized patients (WHO/FAO 2004; Swaminathan and Gerner-Smidt 2007)

Most cases of listeriosis are sporadic. Despite this, foodborne outbreaks due to *L. monocytogenes* have been associated with cheese, raw (unpasteurised) milk, deli meats, salad, fish and smoked fish, ice cream and hotdogs (Montville and Matthews 2005; Swaminathan and Gerner-Smidt 2007) (refer to Table 2).

Table 2: Selected major foodborne outbreaks associated with *L. monocytogenes* (>50 cases and/or ≥1 fatality)

Year	Total no. cases (fatalities)	No. perinatal cases (fatalities)	Food	Country	Comments	Reference
2011	146 (31)	7 (1)	Cantaloupe	US	*Listeria* isolated from cantaloupe and equipment at packing facility, contamination probably occurred in the packing facility	(CDC 2011; FDA 2011)
2009	36 (3)	8 (3)	Chicken wrap	Australia	*Listeria* isolated from pre-packaged chicken wraps, deficiencies in the food safety program for production of chicken meat	(OzFoodNet 2010)
2008	57 (22)	0	Deli meats	Canada	*Listeria* identified on plant equipment, company tried to correct problem with sanitation program; low sodium product	(Government of Canada 2009)
1998–1999	108 (18)	13 (4)	Frankfurters	US	Contamination due to demolition of ceiling refrigeration unit in frankfurter hopper room	(Mead et al. 2006)
1997	1566*	0	Corn and tuna salad	Italy	Possible cross-contamination from other untreated foods	(Aureli et al. 2000)
1992	279 (92)	92 (29)	Jellied pork tongue	France	*Listeria* identified at manufacturing facility	(Norton and Braden 2007)
1985	142 (48)	93 (30)	Mexican-style soft cheese	US	Cheese was made from contaminated milk that was unpasteurised or inadequately pasteurised	(Linnan et al. 1988)
1981	41 (18)	34 (16)	Coleslaw	Canada	Cabbage fertilised with manure from sheep with listeriosis	(Schlech et al. 1983)

* Non-invasive listeriosis

Occurrence in food

L. monocytogenes has been isolated from various ready-to-eat products. In a study by Meldrum et al. (2010) the prevalence of *L. monocytogenes* was 4.1% in crustaceans (n=147), 6.7% in smoked fish (n=178), 2% in sushi (n=50) and 0.9% in green salad (n=335) samples in Wales. Wong et al. (2005) isolated *L. monocytogenes* from 1% of ham (n=104) and 1.7% of pate (n=60) samples in New Zealand. *L. monocytogenes* has also been isolated from dairy products. For example, *L. monocytogenes* was detected in 1.3% of fresh cheese samples in Spain (n=78), 0.2% of hard cheese samples in the United Kingdom (n=1242) and 0.3% of ice creams in Italy (n=1734) (Busani et al. 2005; Cabedo et al. 2008; Little et al. 2009). The prevalence of *L. monocytogenes* in bulk milk tank internationally is 1–60% (FSANZ 2009).

The presence of *L. monocytogenes* in ready-to-eat products is probably due to contamination occurring after the product has been processed. This contamination may occur during additional handling steps such as peeling, slicing and repackaging. Also, in the retail and food service environment, contamination may be transferred between ready-to-eat products (Lianou and Sofos 2007). The type of handling that ready-to-eat meat receives may also influence the level of *L. monocytogenes* contamination. In a survey of retail packaged meats there was a significantly higher prevalence of *L. monocytogenes* reported in products cut into cubes (61.5%) (n=13), compared with sliced products (4.6%) (n=196) (Angelidis and Koutsoumanis 2006).

Host factors that influence disease

People at risk of invasive listeriosis include pregnant women and their foetuses, newborn babies, the elderly and immunocompromised individuals (such as cancer, transplant and HIV/AIDS patients). Less frequently reported, but also at a greater risk, are patients with diabetes, asthma, cirrhosis (liver disease) and ulcerative colitis (inflammatory bowel disease) (FDA 2012).

Dose response

Investigations of foodborne outbreaks of non-invasive listeriosis have concluded that consumption of food with high levels of *L. monocytogenes* (1.9×10^5/g to 1.2×10^9/g) is required to cause illness in the general healthy population (Sim et al. 2002).

The number of *L. monocytogenes* required to cause invasive listeriosis depends on a number of factors. These include the virulence of the particular serotype of *L. monocytogenes*, the general health and immune status of the host, and attributes of the food (for example fatty foods can protect bacteria from stomach acid). Some *L. monocytogenes* serovars are more virulent than others; this may be attributed to differences in the expression of virulence factors which could influence the interactions between the bacterium and the host cells and cellular invasion (Severino et al. 2007). The FDA and WHO have developed separate models for both healthy and susceptible populations to predict the probability that an individual will develop listeriosis (FDA/USDA/CDC 2003; WHO/FAO 2004). The probability that a healthy person of intermediate age will become ill from the consumption of a single *L. monocytogenes* cell was estimated to be 2.37×10^{-14}. For more susceptible populations the probability that illness will occur was estimated to be 1.06×10^{-12}. A more recent assessment on invasive listeriosis in susceptible populations was performed which took into account the different serotypes of *L. monocytogenes* (Chen et al. 2006). This study showed that the probability of a susceptible individual developing invasive listeriosis ranged from 1.31 × 10^{-8} to 5.01 × 10^{-11}, suggesting that there are large differences in virulence between *L. monocytogenes* serotypes.

Recommended reading and useful links

FDA (2012) Bad bug book: Foodborne pathogenic microorganisms and natural toxins handbook, 2nd ed, US Food and Drug Administration, Silver Spring, p. 100–104. http://www.fda.gov/Food/FoodborneIllnessContaminants/CausesOfIllnessBadBugBook/ucm2006773.htm

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