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A LURKING DANGER

To save time and for the sake of convenience, we often reach for ready-to-eat meals, such as pre-made salads. But how safe are they?

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When consumers hear talk of the bacterium *Listeria monocytogenes*, many react with amusement to its challenging pronunciation. However, this species is no laughing matter: it is one of the most dangerous pathogens causing foodborne illnesses. In the first half of 2023, the National Institute of Public Health NIH – Polish Research Institute (PZH-PIB) and the Anti-Epidemic and Border

Sanitation Department of the Chief Sanitary Inspectorate (GIS) recorded 75 cases of listeriosis, which is the disease caused by *Listeria monocytogenes*. Its incidence rate (the number of cases per 100,000 population) is 0.20. In 2022, 46 cases were recorded in the same period, with an incidence rate of 0.12. Although these figures do not seem very high, the disease has a case-fatality rate of 30%.

Characteristics

L. monocytogenes is a rod-shaped bacterium that can thrive with or without oxygen (such organisms are described as “facultative anaerobic”) at temperatures between 20°C and 40°C. However, the bacterium is adapted to variable environmental conditions and can



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– LISTERIOSIS

also survive in temperatures below 20°C. All bacteria of the genus *Listeria* are capable of movement at 25°C. Individual species are distinguished by their ability to ferment sugars and lyse the red blood cells of different types of animals. We currently distinguish 19 species the genus *Listeria*, and only one of them is a human pathogen.

L. monocytogenes has the ability to cross the three most important protective barriers in the human body: the placental, intestinal and blood-brain barriers. The number of infections caused by *L. monocytogenes*, both in Poland and Europe, continues to

grow steadily. Infections occur as isolated cases, but outbreaks of an international scope are also noted.

In adults, the first symptoms occur 24–48 hours after infection. They are initially non-specific and include joint pain, headache, diarrhea, and moderate drowsiness. At higher bacterial concentrations, the infection has acute symptoms: bloody diarrhea and gastroenteritis. Symptoms may resolve spontaneously, but they may further develop into life-threatening listeriosis – especially in high-risk individuals, or YOPI (the young, old, pregnant, immunocompromised).

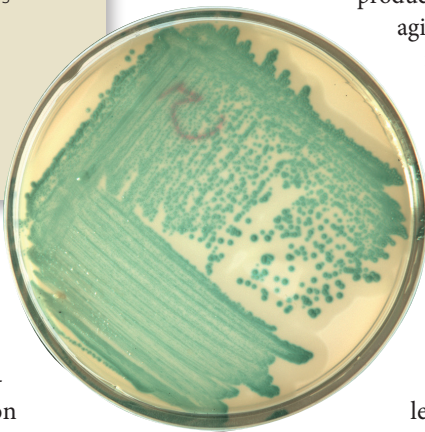
Results of *L. monocytogenes* detection by product type

Product type	Number of contaminated samples	Number of analyzed samples	Share of contaminated samples
Two-ingredient raw salad	1	22	4.5%
Multi-ingredient salads and raw salads	6	38	15.7%
Total	7	60	11.7%

Patients with rapidly-developing listeriosis require hospital care in the following cases:

- organ damage, which may lead to death – as a result of a chronic disease;
- meningitis and encephalitis due to infection of the central nervous system, which is fatal in 50% of cases; importantly, meningitis caused by *L. monocytogenes* is associated with the highest case-fatality rate of all types of bacterial meningitis;
- liver infections caused by sepsis; it mainly affects patients with diabetes, liver cirrhosis, and alcoholism – the case-fatality rate is 50%;
- infection of the endocardium – this type of infection is recorded relatively rarely (7.5%) and mainly affects patients with artificial heart valves;
- listerial conjunctivitis and skin infection – diagnosed mainly in laboratory staff and veterinarians who fail to adhere to proper hygiene regimes.

ALOA® medium (Agar for *Listeria* according to Ottaviani and Agosti) with the growth of *L. monocytogenes* isolated from a raw-vegetable salad. The bluish-green, round colonies are visibly surrounded by an opaque halo in the medium, as a result of phospholipase activity characteristic of the pathogenic *Listeria* species.



The risk of a symptomatic infection and its course depend on the type of contaminated food and on whether the patients belong to any risk group.

Occurrence

The sources of the bacterium include wild and domestic mammals: rodents, sheep, goats, cattle, and pigs. *Listeria* bacilli have also been isolated from birds, fish, and reptiles. Humans can be carriers too – asymptomatic individuals carrying the bacteria are estimated to make up 5–10% of the population. However, more than 95% of all listeriosis cases in humans are

caused by the consumption of contaminated food. The bacterium exhibits special properties that make it an exceptional microbiological hazard in food products. Its optimal growth temperature is 30–37°C, but studies show that it can also survive at temperatures ranging from 3°C to even 40°C. It becomes completely inactivated at temperatures above 75°C. Such a wide temperature range indicates that *L. monocytogenes* is able to survive both what is called low-temperature pasteurization and refrigerated storage, making it a “fridge bacterium.” In addition, it can withstand large fluctuations in pH (from 4.2 to 9.5), drying, and salting. It grows both with and without oxygen and therefore can develop in vacuum-sealed food products and modified atmosphere packaging (MAP). In addition, *L. monocytogenes* is characterized by a high ability to adhere to various surfaces as well as by a low sensitivity to disinfectants.

It is usually isolated from various foods, such as: deli products, smoked fish and fish products, unpasteurized milk and products made from unpasteurized milk, cheese (mainly soft and semi-soft types; less often ripened cheese), frozen food products, ready-to-eat packaged foods, and raw fruits and vegetables.

Prevention

Infection with *L. monocytogenes* poses a high risk and is associated with a high case-fatality rate, so all cases of listeriosis must be reported in accordance with national and international regulations (in Poland, in accordance with the Act on the Prevention and Control of Infections and Infectious Diseases in Humans of 5 December 2008). In the European Union, it is possible

API test confirming the biochemical characteristics of an isolated bacterial colony needed to identify the species



to quickly exchange information on contaminated food and feed through the Rapid Alert System for Food and Feed (RASFF). In addition, food products must be tested for the presence of this bacterium. The criteria for Poland and the European Union are included in Commission Regulation (EC) No 2073/2005 on microbiological criteria for foodstuffs. In ready-to-eat foods for infants and for special medical purposes, *L. monocytogenes* must be absent in 25 g of the product tested, just like in other food products when they leave the manufacturer's facility. In ready-to-eat foods other than foods for infants and for special medical purposes, the product may not exceed 100 cfu/g ("colony forming units"/gram) throughout the shelf life.

L. monocytogenes can grow at low temperatures, so it is important to remember that the average temperatures found on the middle and top shelves in refrigerators, 6–10°C, do not prevent the pathogen from multiplying. More technologically advanced refrigerators offer such features as remote monitoring of temperature and special drawers that maintain the temperature between –3°C and 3°C. This allows safe storage of meat, fish, milk, and cheese. With vegetable products, it is important to maintain a very high level of hygiene, buy only quality products, and maintain "cold chain" continuity on the part of producers, suppliers, and sellers, as well as consumers.

In the Department of Food Hygiene and Quality Management at the Institute of Human Nutrition Sciences, we tested various salads available on the Warsaw market, as well as ready-to-serve vegetable products purchased directly from gastronomic establishments, for the presence of *L. monocytogenes*. The tests were performed using the surface culture technique in keeping with the ISO 11290-1:2017-07 standard. The products were divided into two types: two-ingredient raw salads (containing one or two main ingredients, plus sauce and food additives) and multi-ingredient salads (composed of three or more ingredients in similar or equal proportions by weight, plus sauce and food additives).

L. monocytogenes was detected in a total of seven cases, accounting for 11.7% of all products tested. One of those seven products was a two-ingredient raw salad, while the remaining six products were multi-ingredient salads.

Moreover, the numerical profiles of the *L. monocytogenes* bacteria identified indicate the same strain of bacteria. This suggests that the salads were made from at least one infected ingredient provided by the supplier(s). The detailed ingredients of the seven products that tested positively for *L. monocytogenes* included

Listeriosis in pregnancy

It occurs quite often (accounting for 27% of all cases) and is particularly dangerous.

In this group of infections, we can distinguish three types:

- Maternal listeriosis – it is usually mild (with flu-like symptoms), and some patients are asymptomatic. Since listeriosis poses a high risk in pregnancy (pregnant women are 17 times more susceptible to infection) and may entail serious complications for the mother and the child, the onset of flu-like symptoms alone is enough to suspect listeriosis. Listeriosis in the mother may lead to the fetus becoming infected and consequently to miscarriage.
- Early perinatal listeriosis – intrauterine infection. It is characterized by premature birth. Development of bacteremia in the mother may lead to what is called early infection of the fetus (in the first and second trimesters), which typically results in miscarriage, stillbirth, or preterm labor.
- Late perinatal listeriosis – an infection at a later stage of pregnancy (the third trimester). In this case, the child is born infected with listeriosis. The child may acquire the disease in the womb or during the delivery (passage through the birth canal). A baby carried to full term that becomes infected with *L. monocytogenes* may develop a neurological disease and even die. Fetal and neonatal infections are acute and have a high case-fatality rate. Newborns infected with this bacterium suffer from meningitis, sepsis, and pneumonia.



such additives as meat, fish, cheese, and croutons. Among the infected products, 71% contained meat as one of their ingredients: chicken in four products and ham in one product. Two of the seven infected salads did not contain meat, but they did contain feta cheese and another unspecified type of cheese. All products contained minimally processed (fresh) vegetables. Three products contained a lettuce mix, two products contained Chinese cabbage, and in the case of one sample, Chinese cabbage was the dominant ingredient in terms of weight. Other vegetables found in the list of the ingredients of the infected products included cucumbers and cherry tomatoes.

So what lessons can we formulate at this stage of research? One is that ready-to-eat products should never be stored at a temperature exceeding 4°C. Another is that adding non-plant ingredients to such products is conducive to the occurrence of the pathogen. All in all, this is further evidence that it is simply good practice to buy food products from reliable suppliers. ■

Further reading:

Gurler Z., et al.: The microbiological quality of ready-to-eat salads in Turkey: a focus on *Salmonella* spp. and *Listeria monocytogenes*. *International Journal of Food Microbiology*, 2015, 196: 79–83.

Ferreira V. et al.: *Listeria monocytogenes* Persistence in Food-Associated Environments: Epidemiology, Strain Characteristics and Implications for Public Health. *Journal of Food Protection*, 2014, 77(1): 151–152, 158–160.

Aparecida de Oliveira M. et al.: Microbiological quality of ready-to-eat minimally processed vegetable consumer in Brazil. *Food Control*, 2011, 22: 1400–1403.