

Centre for Enteric Diseases

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Reference: Listeria testing

Listeria monocytogenes

Testing Guidelines for laboratories

Listeriosis is a foodborne illness that may cause very severe disease in pregnant women, neonates, the elderly and immune compromised patients (including HIV and malignancy). It has a very high mortality. Outbreaks have been associated with a wide variety of foods, including dairy products, meat products, vegetables, fruit, and ready-to-eat products.

The causative organism, *Listeria monocytogenes* is a Gram-positive bacillus, that may resemble diphtheroids on Gram stain. *L. monocytogenes* is intrinsically resistant to the cephalosporins and the treatment of choice is ampicillin. Trimethoprim-sulfamethoxazole may be used as alternative treatment in cases of penicillin allergy.

South Africa is currently experiencing a very large outbreak of listeriosis with over 550 cases reported since the start of 2017. As these cases may be missed in the laboratory on examination of Gram stains of blood culture or CSF, this monograph serves as a guide to the laboratory diagnosis of listeriosis in patients. Appropriate clinical indications for testing and guidance for referral of specimens is detailed in Table 1. Please refer suspect food isolates associated with *Listeria* cases as per contacts below, to your nearest public health laboratory*.

	Name	Email	Telephone
Outbreak Response Unit	Dr K McCarthy	kerriganm@nicd.ac.za	011 555-0542
			079 871-7278
Emergency Operations Centre	Mr N Govender	nevashang@nicd.ac.za	011 386-2001
			082 327 2251
Centre for Enteric Diseases	Dr J Thomas	junot@nicd.ac.za	011 555-0426
			073 170-8874
	Dr K Keddy	karenk@nicd.ac.za	011 386-6269
			082 809-5667
	Dr A Smith	anthonys@nicd.ac.za	011 555-0348
	Ms A Sooka	arvindas@nicd.ac.za	011 386-6235
	Ms N Ramalwa	nstienir@nicd.ac.za	011 555-0426
			072 979-9183
*NHLS Infection Control	Dr T Thomas	teena.thomas@nhls.ac.za	011 489-9181
Services Laboratory, Wits			082 740-2897
University Medical School,	Mr R Stewart	rob.stewart@nhls.ac.za	011 489-8578
Hospital Street, Parktown,			011 717-2496
Johannesburg			

Contact information for epidemiological and microbiological support:

Table 1. Clinical indications for testing for listeriosis

Clinical Symptoms/Exposure	Patient Affected	Testing recommended	Where to submit testing
Asymptomatic with consumption of a known contaminated or recalled food product	All patients, including immunocompromised (due to HIV, malignancy, or immunosuppressive medication), pregnant and those ≥65 years of age	 None Counsel regarding symptoms of gastroenteritis and invasive listeriosis Consult a healthcare provider should symptoms develop 	N/A
Gastroenteritis with or without fever and no other symptoms AND	Immunocompetent patients who are NOT pregnant and <65 years of age	 Stool MC&S* Stool culture for <i>Listeria</i> NOT indicated Counsel regarding potential symptoms of invasive listeriosis Evidence of fever within 24 hours of clinical assessment: consider routine blood cultures 	 Routine stool and blood cultures to diagnostic microbiology laboratory – do not culture specifically for Listeria
Consumption of a known contaminated or recalled food product	Immunocompromised, pregnant and those ≥65 years of age	 Stool MC&S Evidence of fever within 24 hours of clinical assessment: consider routine blood cultures Counsel regarding potential symptoms of invasive listeriosis 	 Routine stool and blood cultures to diagnostic microbiology laboratory
Evidence of systemic involvement suggested by headache, stiff neck, confusion, loss of balance Flu-like symptoms (including fever) in immunocompromised persons, pregnant, or ≥65 years of age, particularly when there is a history of preceding gastroenteritis.	All patients	 Routine blood cultures Lumbar puncture for suspected meningitis/meningoencephalitis - CSF MC&S Culture of other potentially affected anatomical sites 	 Routine blood and CSF cultures to diagnostic microbiology laboratory For CSF culture negative specimens, but <u>high index</u> <u>of suspicion</u>, CSF may be submitted to NICD for molecular diagnostics[†] NOTE: CSF from immunosuppressed patients may not contain inflammatory cells, thereby complicating microscopic interpretation

*MC&S: microscopy culture and sensitivity

+For suspected *Listeria* meningitis in culture-negative patients, please contact CED laboratory as per contact details above to arrange for molecular diagnostics on CSF specimens.

Laboratory culture and identification method for Listeria

Specimen type: Cerebrospinal fluid (CSF), Blood culture

Other sterile sites: joint, pleural, pericardial fluid, amniotic fluid and placenta or fetal tissue

Media for specimen processing: 5% blood agar. For CSF, BHI broth may enhance recovery of *Listeria* isolates (Q-Pulse5/docs/active/ MIC1682, MIC1030)

Incubation: aerobic conditions at 37°C for 24 to 48 hours



Figure 1: Clockwise from top left: CSF Gram stain, colony morphology on 5% blood agar, catalase test, colony morphology, tumbling motility using the hanging drop method, Gram stain (1) & (2).



Figure 2. Testing for tumbling motility using the "Hanging drop" method (Q-Pulse5/docs/active/ TADM0150).

Table 2. Differentiating *L. monocytogenes* from other common Gram-positive bacteria that appear similar on blood agar or Gram stain (Q-Pulse5/docs/active/MIC0622v1).

Pathogen	β- haemolysis	Catalase	Motility 25°C	Aesculin	Gram	Growth on 6.5% NaCl
L. monocytogenes	+	+	+	+	GPB	+
S. agalactiae	+	-	-	-	GPC	+
Corynebacterium	-	+	+/-	-	GPB	

