



**NATIONAL INSTITUTE FOR
COMMUNICABLE DISEASES**

Division of the National Health Laboratory Service

DIVISION OF PUBLIC HEALTH SURVEILLANCE AND RESPONSE
NATIONAL INSTITUTE FOR COMMUNICABLE DISEASES (NICD)
NATIONAL HEALTH LABORATORY SERVICE (NHLS)

NICD-NHLS HANDBOOK FOR DIAGNOSIS OF FOODBORNE ILLNESS CLUSTERS / OUTBREAKS

**Developed by:
Division of Public Health Surveillance and Response,
Outbreak Response Unit, NICD-NHLS**

In collaboration with

**Centre for Enteric Diseases, NICD-NHLS
Centre for Emerging and Zoonotic Diseases (Bacteriology), NICD-NHLS
NHLS Public Health Laboratories**

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Related documents

- [NICD Quick Reference Guide to Investigation of Foodborne Disease Outbreaks](#)
- [Foodborne Outbreak Case Investigation form](#)
- [Foodborne Outbreak Line-list](#)
- [NICD-NHLS Centre for Enteric Diseases specimen form](#)
- Specimen request forms
 - [NHLS Infection Control Laboratory Service form](#)
 - [NHLS Public Health Laboratory KZN form](#)
 - [NHLS Public Health Laboratory Port Elizabeth form](#)
- [Suspected botulism case investigation form](#)
- [NICD-NHLS CED specimen collection guide and packaging](#)
- [Quick reference guide for submission of specimens for suspected food/waterborne disease outbreaks to NHLS/NICD laboratories](#)

1. Introduction

Foodborne illness clusters/outbreaks refer to any food-related incident involving 2 or more individuals with onset of illness that is epidemiologically linked to a common food/beverage source, and are notifiable. The cause may be infectious or toxin-related. It is important that all foodborne illness outbreaks be investigated. The attached [Quick Reference Guide to Investigation of Foodborne Disease Outbreaks](#) details the essential steps in investigating such outbreaks. The following guide provides supplementary information for specimen collection and laboratory investigations.

Investigation of food-borne illness has three components: First, clinical symptoms and epidemiological characteristics of the illness should be described. Second, laboratory investigations should be performed on environmental samples (implicated foodstuffs or water) and clinical specimens (vomit, stool or rarely, blood). Third, environmental investigation should be conducted. This document describes the laboratory investigations that are done. Readers are referred elsewhere for resources to investigate the clinical and epidemiological characteristics of the illness.

Depending on the clinical presentation of cases, laboratory investigations should include three aspects: (1) bacterial and bacterial toxin identification, (2) virus identification, and (3) environmental sampling (food, water, and milk testing where applicable). It is recommended that samples of the implicated food/beverage AND clinical samples (i.e. stool, rectal swabs and/or vomit) be collected for all outbreaks when possible, and ALL samples collected should be referred to the designated NHLS Public Health Laboratory (see table under “Contact Details”) unless specified otherwise. These laboratories have the capacities to perform specialised testing for foodborne pathogens and toxins, which may not be routinely detected by standard microscopy and culture techniques.

Specific specimen submission/case investigation forms for foodborne illness incidents/outbreaks should be completed and must accompany specimens. Nevertheless, all specimens should be accompanied by the following minimum set of legible information:

- Patient’s full name and surname, age, gender;
- Name and contact numbers of the attending health worker, and hospital name;
- Name and contact numbers (incl. cell phone) of key investigators (to facilitate communication of results and response);
- A summary of the clinical features of the case(s) and description of the outbreak (e.g. number affected, exposed, and preliminary info on possible source, estimation incubation period).
- Specimen type.
- A clear test request, as outlined in the tables below. (If in doubt, request “Foodborne illness investigation”).
- Label as **“OUTBREAK SPECIMEN”**

2. Bacterial identification (clinical specimens)

2.1 Instructions for healthcare workers

Pathogen, clinical symptoms & comments	Sample collection	Tests to request	Send to	Forms to complete
Bacterial pathogens	Vomiting: vomitus / gastric contents	Foodborne illness investigation	Specimens must be referred to the designated Public Health Laboratory for specialised testing (see contact details below)	Clinical specimens: standard NHLS specimen submission form, clearly stating 'foodborne illness investigation'. Case investigation: Department of Health foodborne illness case investigation forms or foodborne outbreak case investigation form may be used, whichever is available. A line-list should additionally be completed for all investigations.
	Diarrhoea: stool specimen, or rectal swab if stool cannot be obtained If stool specimen cannot be processed within 2 hours: use a cotton-tipped swab to sample stool specimen. Place swab with sampled stool in Cary-Blair transport medium. Submit both the initial container with stool and swab in transport medium to the laboratory. Transport refrigerated, do NOT freeze.			
	Fever: blood culture and serum			
	Environmental isolates in water/food-borne disease outbreaks only.	Suspected botulism	Specimens must be referred to the Centre for Emerging and Zoonotic Diseases (Bacteriology), NICD-NHLS (see contact details below)	Suspected botulism: Suspected botulism case investigation form must be used
Neurological* features: serum, stool and vomitus/gastric contents PLUS any suspected food items *Descending motor nerve paralysis without sensory changes, including cranial nerve palsies. NB. There may be initial gastrointestinal symptoms e.g. nausea, vomiting, with or without diarrhoea				

When enteric pathogens (such as *Salmonella* spp.) are cultured, isolates are referred to the Centre for Enteric Diseases (Bacteriology), NICD for further characterisation. This will enable the detection of widespread foodborne illness outbreaks, in addition to fully characterising local outbreaks. Enteric pathogens submitted to Centre for Enteric Diseases should indicate outbreak isolates.

2.2 Laboratory test information

Available tests	Special instructions	Testing laboratory
Microscopy, culture and antimicrobial susceptibility (MC&S) and bacterial toxin detection	Transport refrigerated, do NOT freeze.	Designated NHLS Public Health Laboratory
Diarrhoeagenic <i>E. coli</i> : identification, extended serotyping, virulence gene identification via multiplex PCR, DNA fingerprinting of strains via PFGE analyses	Isolates sub-cultured onto Dorset transport medium incubated at 37°C overnight at source laboratory, prior to submission to Centre for Enteric Diseases (Bacteriology), NICD-NHLS	Centre for Enteric Diseases (Bacteriology), NICD-NHLS 011-555-0333/4
<i>Vibrio</i> species, <i>Salmonella</i> and <i>Shigella</i> : identification, extended serotyping, DNA fingerprinting of strains via PFGE analyses and MLVA.	Isolates sub-cultured onto Dorset transport medium incubated at 37°C overnight at source laboratory, prior to submission to Centre for Enteric Diseases (Bacteriology), NICD-NHLS	Centre for Enteric Diseases (Bacteriology), NICD-NHLS 011-555-0333/4
<i>Listeria</i> and <i>Campylobacter</i> : Identification, genotyping (MultiLocus Sequence Typing [MLST])	Isolates sub-cultured onto Dorset transport medium incubated at 37°C overnight at source laboratory, prior to submission to Centre for Enteric Diseases (Bacteriology), NICD-NHLS	Centre for Enteric Diseases (Bacteriology), NICD-NHLS 011-555-0333/4
Botulism testing	Transport refrigerated, do NOT freeze.	Centre for Emerging and Zoonotic Diseases, Special Bacterial Pathogens Reference Laboratory, NICD-NHLS 011-555-0306/31

3. Viral identification

Tests for viral pathogens that may be the cause of foodborne illness incidents are not routinely performed, and these need to be requested specifically through liaison and discussion with the Centre for Enteric Diseases (Virology) and the Outbreak Response Unit, NHLS-NICD.

3.1 Instructions for Healthcare Workers:

Pathogen, clinical symptoms & comments	Sample collection	Tests to request	Send to	Forms to complete
Viral pathogens	Stool (note, rectal swabs are unsuitable specimens)	Viral gastroenteritis	Centre for Enteric Diseases (Virology), NICD- NHLS. 1 Modderfontein Rd., Sandringham, Gauteng, 2192.	NICD-NHLS Centre for Enteric Diseases specimen form

3.2 Laboratory test information:

Available tests	Special Instructions	Testing laboratory
Rotavirus, norovirus, sapovirus, adenovirus, astrovirus, Taqman array screening (screen for range of viral, bacterial and parasitic pathogens)	Store and transport at 4°C. Maintenance of cold chain is recommended from collection to receipt at NICD	Centre for Enteric Diseases (Virology), NICD-NHLS. 011-555-0370

4. Environmental sampling: food, milk and water testing

Contact the public health laboratory for instructions on specimen collection and submission. Sampling should be systematic, and should cover as wide a range as possible. Different types of foods must be placed in separate containers. The quantity of food taken must be sufficient. An environmental health practitioner must notify the owner/manager at the food production/catering facility that samples need to be taken (provide a reason). Samples for microbiological analysis must be collected with decontaminated equipment (immerse equipment in 70% alcohol and flame using a portable spirit burner until alcohol evaporates, allow cooling before using).

4.1 Instructions for Environmental Health Practitioners (and other health authorities):

Pathogen, clinical symptoms & comments	Sample collection	Tests to request	Send to	Forms to complete
Food- and water-borne pathogens	Food: Vol: 50-100g (container full); Container: sterile; Transport temp: 2 - 8 deg C Rejection criteria: >48 hours at room temperature it should be refrigerated, container not sterile, unwholesome	Liaise with testing laboratory	Designated NHLS public health laboratory	Designated public health laboratory submission form (links to individual available under 'Related documents' at the beginning of this guideline)
	Milk: Vol: 100ml; Container: original/ sterile; Transport temp: 2 - 8 deg C Rejection criteria: >48 hours old, sour, frozen, container not sterile, at room temperature			
	Water (potability): Vol: 100-200ml; Container: sterile; Transport temp: 2 - 8 deg C Rejection criteria: >24 hours at room temperature			
	Water (<i>Salmonella</i> spp., <i>V. cholerae</i>, etc.): Vol: moore pad in sterile container, 1000ml for each test; Container: clean / new / sterile; Transport temp: room temperature (if delivered the same day) Rejection criteria: >24 hours, unsterile container, 48 hours at room temperature			

4.2 Testing of clinical and environmental specimens by specialised laboratories for food- and waterborne outbreaks

Laboratory	NHLS-ICSL		NHLS Public Health, Port Elizabeth		NHLS Public Health, KZN	
	Clinical ¹	Env ²	Clinical ¹	Env ²	Clinical ¹	Env ²
<i>Salmonella</i> spp.	✓	✓	✓	✓	✓	✓
<i>Shigella</i> spp.	✓	✓	✓	✓	✓	✓
<i>E. coli</i> O157	✓	✓	x	✓	✓	✓
<i>Bacillus cereus</i>	✓	✓	✓	✓	✓	✓
<i>Clostridium perfringens</i>	✓	✓	✓	✓	✓	✓
<i>Campylobacter</i> spp.	✓	✓	x	✓	✓	✓
<i>Listeria monocytogenes</i>	✓	✓	x	✓	✓	✓
<i>Staphylococcus aureus</i>	✓	✓	✓	✓	✓	✓
<i>Vibrio</i> spp.	✓	✓	✓	✓	✓	✓
Bacterial toxins ³	✓	✓	x	✓	x	x

¹Clinical specimens: stool, vomitus.

²Environmental specimens: food, water, milk.

³*C. perfringens* enterotoxin from stools and isolates, *S. aureus* and *B. cereus* enterotoxin tests from isolates, NOT botulinum toxin testing, which is only done at NICD (see 2.1 above).

5. Contact Details

It is essential for public health officials / healthcare workers investigating foodborne illness outbreaks to notify the NHLS staff when specimens are collected as part of an outbreak. ALL specimens collected should be referred to one of the designated public health laboratories unless specified otherwise:

5.1 Designated NHLS Public Health Laboratories contact details and tests offered

	*Provinces referring to designated public health laboratory	Laboratory	Address	Contact Person	Tests offered
1	Gauteng Limpopo Mpumalanga North West Northern Cape Free State Western Cape	NHLS Infection Control Service Laboratory (ICSL), Johannesburg	Wits Medical School, Room 3T09, 7 York Rd. Parktown, Johannesburg, 2193.	Mr. Rob Stewart Tel. 011-489-8578/9 or 011-717-2496 Dr Teena Thomas Tel. 011-489-9181	Foodborne pathogen tests offered as follows: <ul style="list-style-type: none"> • <i>Salmonella</i> spp. • <i>Shigella</i> spp. • <i>E. coli</i> 0157 • <i>Bacillus cereus</i> • <i>Clostridium perfringens</i> • <i>Campylobacter</i> spp. • <i>Listeria monocytogenes</i> • <i>Staphylococcus aureus</i> • <i>Vibrio</i> spp. • **Bacterial toxins
2	KwaZulu-Natal (KZN)	NHLS Public Health Laboratory KZN	3rd Floor, 149 Prince Street, Durban, 4001.	Ms Esther Pillay Tel: 031-327-6743	
3	Eastern Cape	NHLS Public Health Laboratory, Port Elizabeth (PE)	Corner of Buckingham and Eastborne Road, Mount Croix, Port Elizabeth, 6000.	Ms Vanessa Pearce Tel: 041-395-6174	KZN: Confirmation of <i>Campylobacter</i> spp. referred to NHLS Addington Microbiology Laboratory ***Testing for bacterial toxins not offered – referred to NHLS-ICSL. - PE: *** Testing for <i>E. coli</i> O157, <i>Campylobacter</i> spp., <i>Listeria monocytogenes</i> , & bacterial toxins is referred to NHLS-ICSL.

*Samples/specimens may also be transported directly to the NHLS-ICSL for testing; **Bacterial toxins: *Bacillus cereus*; *Staphylococcus aureus*; *Clostridium perfringens*; ***Public health laboratories to refer to NHLS-ICSL, Johannesburg

Healthcare workers and laboratory staff may additionally utilise the following contact points for discussion of cases/outbreaks if needed:

- **NICD Hotline:** 082-883-9920 (for use by healthcare professionals only)
- **NICD Outbreak Response Unit, Division of Public Health Surveillance and Response:** outbreak@nicd.ac.za or 011-555-0542/0395
- **NHLS Infection Control Services Laboratory (ICSL):** 011-489-8578/9 or 011-717-2496, rob.stewart@nhls.ac.za or Dr Thomas: 011-489-9181
- **NHLS Public Health Laboratory KZN :** Ms Esther Pillay: 031-327-6743, esther.pillay@nhls.ac.za
- **NHLS Public Health Laboratory Port Elizabeth:** Ms Vanessa Pearce: 041-395-6174, vanessa.pearce@nhls.ac.za

6. Procedure to follow when NHLS laboratories receive specimens for foodborne illness investigations

1. All environmental samples (e.g. food, water etc.) and clinical specimens (with accompanying requests forms) related to foodborne illness incidents/outbreaks that are received at the NHLS diagnostic laboratories should be forwarded as soon as possible to one of the designated public health laboratories as indicated above. Both clinical and environmental samples can be tested at these (public health) laboratories. Routine MC&S will not detect foodborne pathogens. When botulism is suspected, specimens should be referred to the Centre for Emerging and Zoonotic Diseases (CEZD-Bacteriology) at the NICD, but only after consulting CEZD and the Outbreak Response Unit. When viral pathogens testing are requested as part of foodborne disease outbreak investigations, this should be discussed with the Centre for Enteric Diseases (Virology) and the Outbreak Response Unit, NICD-NHLS, as these tests are not done routinely.
2. Laboratory personnel must contact the designated public health laboratory to inform staff that samples/specimens will be forwarded to their laboratory for testing.
3. If MC&S had been performed on clinical specimens (e.g. stools) at the local laboratory, and it was established afterwards that the specimen/s are part of a foodborne illness incident/outbreak – if these specimens are still available, they may still be sent to the designated public health laboratory for further testing. Please discuss with the public health laboratory.

7. Procedure to follow when NHLS Public Health Laboratories receive specimens for foodborne illness investigations

1. Once specimens are received at the public health laboratory, inform the Outbreak Response Unit, NICD via e-mail/phone, preferably through forwarding the request/submission form, in order for the Unit members to follow-up the incident.
2. Once the results of investigations are available, the Public health laboratory is requested to alert the Outbreak Response Unit and the specific Department of Health officials, and the referring NHLS laboratory which submitted the specimen. The Outbreak Response Unit will simultaneously communicate results to the outbreak investigation team in the field and other relevant stakeholders.
3. When enteric bacterial pathogens (such as *Salmonella* spp.) are cultured, we request that the Public Health laboratory refer the isolates to the Centre for Enteric Diseases (CED) - Bacteriology, NICD for further characterisation. (CED- Bacteriology - Tel. 011-386-6235 or 011-555-0333). CED staff will alert ORU of the laboratory results
4. When a viral pathogen (e.g. a norovirus) is clinically suspected as the cause of a foodborne disease outbreak, testing of limited number of stool specimens only (rectal swabs are unsuitable specimens) can be done by the Centre for Enteric Diseases (Virology), NICD (Tel: 011-555-0370 or 011-386-6549) with prior arrangement. This will be decided on a case-by-case basis in consultation with Outbreak Response Unit and the Centre for Enteric Diseases (Virology).