Guide to Laboratory Services: Microbiology

Arizona Department of Health Services
Bureau of State Laboratory Services
250 North 17th Avenue
Phoenix, Arizona 85007
(602) 542-1188

05.09.2019

Victor Waddell Ph.D.
Bureau Chief

Daniel M. Lavine, M.D.
Laboratory Director

Linda Getsinger
Assistant Bureau Chief
Table of Contents

Table of Contents ............................................................................................................................................. 1
General Information........................................................................................................................................... 1
Arizona State Public Health Laboratory Contact Information ................................................................. 2
Core Functions and Capabilities of State Public Health Laboratories ...................................................... 3
Specimen Rejection Policy............................................................................................................................... 4
Directory of Laboratory Services ................................................................................................................ 5
Section 1: Bacteriology..................................................................................................................................... 9
  Botulism ........................................................................................................................................................ 11
  Carbapenem Resistance Testing ................................................................................................................ 12
  Culture Independent Diagnostic Test (CIDTs) Isolate Recovery .............................................................. 13
  Diphtheria .................................................................................................................................................... 13
  Enteric Culture .......................................................................................................................................... 14
  Haemophilus ............................................................................................................................................... 15
  Legionella ................................................................................................................................................ 16
  Leptospirosis ............................................................................................................................................ 17
  Listeria ....................................................................................................................................................... 18
  Meningococcus ......................................................................................................................................... 19
  Pertussis ................................................................................................................................................... 20
  Shigella ..................................................................................................................................................... 21
  Shiga Toxigenic E. coli ............................................................................................................................... 22
  VISA / VRSA ............................................................................................................................................ 23
  Vibrio ......................................................................................................................................................... 24
Section 2: Mycobacteriology ........................................................................................................................... 25
Section 3: Limited Parasitology ........................................................................................................................ 29
Section 4: Serology .......................................................................................................................................... 32
Section 5: Virology .......................................................................................................................................... 38
  Rabies ......................................................................................................................................................... 44
Section 6: Foodborne Pathogens ..................................................................................................................... 46
  Clinical Testing .......................................................................................................................................... 46
  Food Product Samples .............................................................................................................................. 48
  Water Samples ......................................................................................................................................... 50
Section 7: Bioemergency Detection and Response ....................................................................................... 52
  Anthrax ..................................................................................................................................................... 55
  Brucellosis ............................................................................................................................................... 56
  Ebola ......................................................................................................................................................... 57
  Middle East Respiratory Virus, MERS-CoV ............................................................................................ 58
  Melioidosis and Glanders ......................................................................................................................... 59
  Orthopoxvirus ......................................................................................................................................... 60
  Plague ....................................................................................................................................................... 62
  Q Fever .................................................................................................................................................... 63
  Tularemia .................................................................................................................................................. 64
Section 8: Sample Submission Guidelines .................................................................................................... 65
  IATA Category A Organisms .................................................................................................................. 69
  Category A Shipping Examples ................................................................................................................ 70
  Category B Shipping Examples ................................................................................................................ 71
  Category B Checklist ............................................................................................................................... 72
Section 9: Requesting Collection Kits and Mailing Containers ...................................................................... 74
General Information

Bureau Chief, Laboratory Services    Victor Waddell Ph.D.
Director, Laboratory Services     Daniel M. Lavine, M.D.
Assistant Bureau Chief      Linda Getsinger

Hours of Operation: 8:00 AM to 5:00 PM Monday through Friday (Emergency services available on nights or weekends when required by public health needs.)

Receiving section only is open from 9:30am to 4:30pm on Saturday

Annual Holiday Schedule: Laboratory Services observes all state recognized holidays.

Location: 250 North 17th Avenue, Phoenix, Arizona 85007

Telephone Number: (602) 542-1188
WATTS Line: (800) 525-8915
Fax Number: (602) 542-0760

Emergency Phone (Weekends/After Hours): (480) 303-1676
# Arizona State Public Health Laboratory Contact Information

<table>
<thead>
<tr>
<th>Section</th>
<th>Supervisor</th>
<th>Contact information</th>
</tr>
</thead>
<tbody>
<tr>
<td>Receiving /Shipping</td>
<td>Jennifer Elston</td>
<td><a href="mailto:labreceiving@azdhs.gov">labreceiving@azdhs.gov</a> or (602) 542-1190</td>
</tr>
<tr>
<td>Chief Microbiology</td>
<td>Stacy White, Ph.D.</td>
<td>(602) 542-6146</td>
</tr>
<tr>
<td>Bioemergency Response and Detection for</td>
<td></td>
<td>(602) 364-0999</td>
</tr>
<tr>
<td>Select Agents</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Virology/Serology</td>
<td>Kathryn Fitzpatrick</td>
<td>(602) 542-0968</td>
</tr>
<tr>
<td>TB/Mycobacteriology</td>
<td>Drew Francis</td>
<td>(602) 542-6132</td>
</tr>
<tr>
<td>Bacteriology / (Limited) Parasitology</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Foodborne Pathogens</td>
<td>Roumen Penev, Ph.D.</td>
<td>(602) 542-6130</td>
</tr>
<tr>
<td>Analytical Preparation Services</td>
<td>Michael Worsham</td>
<td>(602) 542-1147</td>
</tr>
</tbody>
</table>
Core Functions and Capabilities of State Public Health Laboratories

State public health laboratories face the broad challenge of working towards prevention and control of disease and improvement of health. To function in this capacity, the public health labs provide testing for, and aid in the diagnosis of, unusual pathogens. The labs serve as the first line of defense in the rapid recognition and prevention of the spread of communicable diseases, while also serving as centers of expertise for the detection and identification of biologic agents of importance in human disease. The public health labs also perform testing to meet the specific program needs of the public health agencies.

Routine diagnostic testing for hospitals and private laboratories is provided through independent reference laboratories.

The policy of the Arizona State Public Health Laboratory (ASPHL) is to provide microbiology and immunology diagnostic support to federal, state, county, and tribal agencies. In addition, the ASPHL serves as a reference microbiology laboratory to hospital and independent clinical laboratories in order to confirm their atypical results from cultures and clinical specimens. This information is also used as part of the Department of Health Services disease surveillance program. Selected diagnostic test procedures are available to private medical practitioners when a procedure is not available through independent reference laboratories or when intense surveillance is deemed necessary. The laboratory also accepts food and water from county and state agencies for outbreak investigations and surveillance. Upon specific request to the State Laboratory, the ordering provider (listed in the Ordering Provider Information Section of the Microbiology Sample Submission form) will be given test results in addition to the submitting agency. Otherwise, test results will be only given to the submitting agency.

ASPHL reporting requirements can be found at http://azdhs.gov/labreporting. This report identifies agents which must be reported to the state and which isolates must be submitted to the laboratory. Please follow packing guidelines found in this manual, or on our website at http://www.azdhs.gov/lab/

The ASPHL provides specimen collection materials and mailers free of charge. Further information regarding specimen collection materials, mailing containers and Request for Materials Form is located in Section 09: Requesting Collection Kits and Mailing Containers. All requisitions and supplies for specimen submission are available through the Receiving Section in Phoenix at http://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving

The purpose of this manual is to provide a ready reference to our clients and to assist them in obtaining laboratory services as efficiently as possible. Charts are provided for quick reference and more detailed information is available by test or organism name in each section of the manual. This manual can be downloaded or viewed at http://www.azdhs.gov/lab/documents/microbiology/lab-guide.pdf

Issued: 05.09.2019  Controlled Copy # Uncontrolled
Specimen Rejection Policy

The ASPHL currently has the following policy for rejection of laboratory specimens and/or requested examinations. The ASPHL will NOT examine clinical/reference specimens if the following circumstances exist:

- Test is routinely available at a hospital or a private independent laboratory.
- The identifier on the specimen did not match the identifier on the submission form, or there was no identification on the specimen.
- The quantity of specimen was not sufficient for examination.
- The specimen was too long in transit between the time of collection and receipt in the laboratory.
- The specimen was broken or leaked in transit.
- Clinical/epidemiological information submitted with the specimen was either insufficient or incomplete.
- Specimen was submitted in an improper or expired container, transport media or preservative.
- Blood specimens were hemolyzed or contaminated.
- Only acute blood specimen was submitted, no convalescent specimen (if applicable)
- Material for rabies examination was too decomposed or desiccated to test.
- Reference cultures were mixed or contaminated; only pure cultures are acceptable.
- Tissues were not submitted in individual containers.
- Test request deemed unnecessary by the Bureau of Epidemiology and Disease Control.

Exceptions to this policy will be considered due to extenuating circumstances; however, final approval to make an exception can only be made by the Laboratory Director, Bureau Chief, Assistant Bureau Chief, or Technical Supervisor.
# Directory of Laboratory Services

The following table lists the diagnostic and reference services offered by the Office of Public Health Microbiology. The table is organized alphabetically by disease or agent for easy reference. Please go to the specified laboratory section of this manual for more detailed information on collection and submission of laboratory samples.

<table>
<thead>
<tr>
<th>Disease or Agent</th>
<th>Culture</th>
<th>Detection</th>
<th>Toxin</th>
<th>Smear</th>
<th>Serology Single Serum</th>
<th>Serology Paired Sera</th>
<th>Laboratory Section</th>
<th>Manual Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adenovirus</td>
<td></td>
<td>PCR***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Anthrax</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td><em>Bacillus anthracis</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bacillus cereus</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Foodborne Pathogens</td>
<td>Section 6</td>
</tr>
<tr>
<td>Blood/Tissue Parasites</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td>Parasitology</td>
<td>Section 3</td>
</tr>
<tr>
<td>Botulism</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Referred to CDC</td>
</tr>
<tr>
<td><em>Clostridium botulinum</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Referred to CDC</td>
<td>Section 1</td>
</tr>
<tr>
<td>Brucella</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td><em>Burkholderia mallei/pseudomallei</em></td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td><em>Campylobacter</em></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Chikungunya</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Serology</td>
<td>Virology</td>
</tr>
<tr>
<td>Carbapenem resistant Enterobacteriaceae (CRE)/Pseudomonas aeruginosa (CRPA)</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Culture independent test (CIDT) isolate recovery</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Foodborne Pathogens</td>
</tr>
<tr>
<td>Dengue</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Serology</td>
<td>Virology</td>
</tr>
<tr>
<td>Diphtheria <em>Corynebacterium diphtheriae</em></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Disease or Agent</td>
<td>Culture</td>
<td>Detection</td>
<td>Toxin</td>
<td>Smear</td>
<td>Serology Single Serum</td>
<td>Serology Paired Sera</td>
<td>Laboratory Section</td>
<td>Manual Location</td>
</tr>
<tr>
<td>----------------------------------</td>
<td>---------</td>
<td>-----------</td>
<td>---------</td>
<td>-------</td>
<td>-----------------------</td>
<td>----------------------</td>
<td>--------------------</td>
<td>------------------</td>
</tr>
<tr>
<td>Ebola</td>
<td></td>
<td>PCR</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>E. coli</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Foodborne Pathogens</td>
<td>Section 6</td>
</tr>
<tr>
<td>Enterovirus</td>
<td>-</td>
<td>PCR***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Hantavirus</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Serology Referred to CDC</td>
<td>Section 4</td>
</tr>
<tr>
<td>Haemophilus</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>HIV</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Human Metapneumovirus</td>
<td>-</td>
<td>PCR***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Influenza</td>
<td>-</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Intestinal Parasites (Giardia and Cryptosporidium)</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Limited Parasitology</td>
<td>Section 3</td>
</tr>
<tr>
<td>Legionella</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Legionella</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Listeria</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Lyme Disease</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Serology Referred to CDC</td>
<td>Section 6</td>
</tr>
<tr>
<td>Borrelia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Measles</td>
<td>-</td>
<td>PCR</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Virology Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Neisseria meningitidis</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>MERS-CoV</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>Mumps</td>
<td>-</td>
<td>PCR</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Virology Serology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Murine Typhus</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Serology Referred to CDC</td>
<td>Section 4</td>
</tr>
<tr>
<td>Rickettsia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mycobacterium</td>
<td>X</td>
<td>NAAT PCR</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Mycobacteriology</td>
<td>Section 2</td>
</tr>
<tr>
<td>Disease or Agent</td>
<td>Culture</td>
<td>Detection</td>
<td>Toxin</td>
<td>Smear</td>
<td>Serology Single Serum</td>
<td>Serology Paired Sera</td>
<td>Laboratory Section</td>
<td>Manual Location</td>
</tr>
<tr>
<td>-----------------</td>
<td>---------</td>
<td>-----------</td>
<td>-------</td>
<td>-------</td>
<td>-----------------------</td>
<td>---------------------</td>
<td>--------------------</td>
<td>-----------------</td>
</tr>
<tr>
<td>Norovirus</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Orthopoxvirus</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>Parainfluenza viruses (1-4)</td>
<td></td>
<td>PCR***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Pertussis</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Bordetella pertussis</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Q Fever</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>Coxiella burnetii</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Rabies</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Rocky Mountain Spotted Fever</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Rickettsia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Respiratory Syncytial Virus (RSV)</td>
<td></td>
<td>X***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Rhinovirus</td>
<td></td>
<td>X***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Virology</td>
<td>Section 5</td>
</tr>
<tr>
<td>Rubella</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Salmonella</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Foodborne Pathogens</td>
<td>Section 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Section 6</td>
</tr>
<tr>
<td>Shiga toxin - producing E. coli</td>
<td></td>
<td>X</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*Shigella (For identification and confirmation only)</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Foodborne Pathogens</td>
<td>Section 6</td>
</tr>
<tr>
<td>St. Louis Encephalitis</td>
<td></td>
<td>PCR**</td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Staphylococcus (VISA/VRSA)</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Tularemia</td>
<td></td>
<td>X</td>
<td></td>
<td>PCR</td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>Francisella tularensis</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Varicella-Zoster</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Referred to CADPH</td>
<td>Section 5</td>
</tr>
<tr>
<td>Vibrio</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Foodborne Pathogens</td>
<td>Section 6</td>
</tr>
<tr>
<td>Disease or Agent</td>
<td>Culture</td>
<td>Detection</td>
<td>Toxin</td>
<td>Smear</td>
<td>Serology Single Serum</td>
<td>Serology Paired Sera</td>
<td>Laboratory Section</td>
<td>Manual Location</td>
</tr>
<tr>
<td>------------------------</td>
<td>---------</td>
<td>-----------</td>
<td>-------</td>
<td>-------</td>
<td>-----------------------</td>
<td>----------------------</td>
<td>-------------------------------</td>
<td>-----------------</td>
</tr>
<tr>
<td>West Nile</td>
<td></td>
<td>PCR**</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
<tr>
<td>Yersinia pestis</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bioemergency</td>
<td>Section 7</td>
</tr>
<tr>
<td>Yersinia (non-pestis)</td>
<td>X</td>
<td>PCR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriology</td>
<td>Section 1</td>
</tr>
<tr>
<td>Zika virus</td>
<td></td>
<td>PCR</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>Serology</td>
<td>Section 4</td>
</tr>
</tbody>
</table>

*Reference isolates no longer accepted.

NAAT = Nucleic Acid Amplification Test
PCR = Polymerase Chain Reaction
PCR** = on mosquito pools only
*** On Approval Only

Specimens should be forwarded to the Centers for Disease Control and Prevention (CDC), California Department of Public Health (CADPH), or Minnesota Department of Public Health (MDH) from the ASPHL and should not be sent directly from other facilities; please contact the appropriate section within the ASPHL with any questions or for sample coordination.
Upon receipt at the ASPHL, all specimens are logged in and assigned to the appropriate area for processing. The time required to process a microbiology specimen varies considerably, as indicated by the following table. Detailed information on the collection and submission of laboratory samples on any of the following tests can be obtained in the following narrative guidelines.

During outbreaks, the Bureau of Epidemiology and Disease Control may conduct surveillance to determine the extent of the outbreak or to determine the relatedness of microorganisms identified in the outbreak. The Office of Public Health Microbiology will support these outbreak investigations through the use of various tools. Data may be shared in these investigations with other states and the CDC in the event of a multi-state outbreak.

For testing assistance for organisms/diseases not offered by the ASPHL please refer to the Center for Disease Control and Prevention (CDC) website (http://www.cdc.gov/laboratory/specimen-submission/list.html) or Minnesota Public Health Laboratory (MPHL). Information regarding available tests, appropriate specimen type(s), collection and storage conditions, shipping requirements and appropriate test authorizations can be found on this website. Specimens should be forwarded to the CDC from the ASPHL and should not be sent directly from other facilities; please contact the Bacteriology section with any questions or for sample coordination with bacteriology or parasitology related samples.

<table>
<thead>
<tr>
<th>Organism/Disease</th>
<th>Specimen</th>
<th>Transport Medium</th>
<th>Comments</th>
<th>Turn Around Time (TAT) Business Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Botulism <em>Clostridium botulinum</em></td>
<td>Serum, Feces, Food</td>
<td>None</td>
<td>Testing requires prior approval by Epidemiology and Disease Control</td>
<td>Referred to CDC</td>
</tr>
<tr>
<td><em>Campylobacter</em></td>
<td>Feces</td>
<td>Cary-Blair or Modified Cary-Blair</td>
<td>Reference isolates no longer accepted. See Enteric Culture</td>
<td>3-5 days</td>
</tr>
<tr>
<td>Carbapenem resistant testing Enterobacteriaceae (CRE)/Pseudomonas aeruginosa (CRPA)**</td>
<td>Pure isolate</td>
<td>Culture Plate/Slant</td>
<td>Submit antimicrobial resistance test results along with isolate</td>
<td>7-10 days</td>
</tr>
<tr>
<td><em>Diphtheria</em> <em>Corynebacterium diphtheriae</em></td>
<td>Throat (membrane) or NP Swab</td>
<td>Semi-sold transport media</td>
<td>Call before submitting</td>
<td>4-6 days</td>
</tr>
<tr>
<td>CIDT isolate recovery</td>
<td></td>
<td></td>
<td>See Enteric culture</td>
<td></td>
</tr>
<tr>
<td>Organism/Disease</td>
<td>Specimen</td>
<td>Transport Medium</td>
<td>Comments</td>
<td>Turn Around Time (TAT) Business Days</td>
</tr>
<tr>
<td>------------------</td>
<td>----------</td>
<td>------------------</td>
<td>----------</td>
<td>-------------------------------------</td>
</tr>
<tr>
<td>Enteric Culture</td>
<td>Feces in transport media</td>
<td>Cary-Blair, or Modified Cary-Blair</td>
<td></td>
<td>5-9 days</td>
</tr>
<tr>
<td><em>Haemophilus</em> **</td>
<td>Pure Culture of Isolate from Sterile Body Site</td>
<td>Chocolate agar slants*</td>
<td>Children 5 years and younger</td>
<td>9-15 days; non B serogroups sent to MDH</td>
</tr>
<tr>
<td>Legionella</td>
<td>Pure Culture of Isolate</td>
<td>BCYE slant or plate</td>
<td>Transport samples at 4°C (cold packs)</td>
<td>Preliminary 3-4 days; Sent to CDC for identification/serotyping</td>
</tr>
<tr>
<td><em>Leptospira</em></td>
<td>Blood with Heparin, CSF, Urine</td>
<td>Fletchers Media</td>
<td>Transport at 20 °C – 30 °C</td>
<td>Preliminary 3-4 days; Sent to CDC</td>
</tr>
<tr>
<td><em>Listeria</em> **</td>
<td>Pure Culture of Isolate</td>
<td>Blood agar/or Agar slant</td>
<td>Ship at 4 °C</td>
<td>Preliminary 4-8 days; Sent to CDC for serotyping</td>
</tr>
<tr>
<td><em>Neisseria meningitidis</em> **</td>
<td>Pure Culture of Isolate from Sterile Body Site</td>
<td>Chocolate agar/or Chocolate Agar slant</td>
<td>Do not refrigerate</td>
<td>Sent to MDH for serotyping</td>
</tr>
<tr>
<td>Pertussis <em>Bordetella pertussis</em></td>
<td>Nasopharyngeal Swab or isolate</td>
<td>Reagan-Lowe</td>
<td>Use polyester NP swabs</td>
<td>PCR 7 days</td>
</tr>
<tr>
<td>VISA/VRSA <em>Staphylococcus</em> **</td>
<td>Pure Culture of Isolate</td>
<td>Culture Plate/Slant</td>
<td></td>
<td>5-7 days</td>
</tr>
<tr>
<td><em>Vibrio</em></td>
<td>Pure Culture of Isolate</td>
<td>Culture Plate/Slant</td>
<td>Do not refrigerate</td>
<td>4-6 days</td>
</tr>
</tbody>
</table>

** Reference cultures only
Botulism

_Clostridium botulinum_

All Botulinum testing for patient specimens is performed at the CDC. Approval for botulism testing must be obtained from the Bureau of Epidemiology and Disease Control prior to submission. Contact the Infectious Disease Section of the Bureau of Epidemiology and Disease Control at (602) 364-3676 (main number)/ (480) 303-1191 (after hours number).

Refer to the Collection section below for appropriate sample types.

**Collection**

**Infant Botulism**
1. Stool for culture and toxin – 10 to 20 grams (or as much as possible)
   - If an enema is needed, use sterile non-bacteriostatic water
2. Rectal swab
3. Food or other potential sources for toxin and culture

**Foodborne Botulism – Adult**
1. Serum – 5 to 15 mL without anticoagulant (serum samples must be collected before antitoxin treatment)
2. Stool – 10 to 20 grams (If an enema is needed, use sterile non-bacteriostatic water)
3. Vomitus
4. Gastric contents
5. Remainder of suspected food

**Wound Botulism**
1. Serum – 5 to 15 mL (serum samples must be collected before antitoxin treatment)
2. Debrided tissue, exudate or swab samples from wound
3. Stool 10 to 20 grams (to rule out foodborne botulism)

**Shipment of Specimens**

All specimens should be kept at refrigerated temperatures during storage and shipment. Shipment should contain ice or cool packs.

See Section 8: Sample Submission Guidelines.

All specimens will be forwarded on to the Centers for Disease Control in Atlanta, Georgia for testing.

**Reporting and Interpretation of Results**

The State Laboratory will notify the submitting agency and the Bureau of Epidemiology and Disease Control with results of the botulism testing as soon as they are available.
Section 1: Bacteriology

Carbapenem Resistance Testing

*Enterobacteriaceae* and *Pseudomonas aeruginosa* isolates demonstrating Carbapenem resistance may be submitted for additional characterization. Carbapenemase activity of an organism is determined by using the modified Carbapenem Inactivation Method (mCIM). Suspect isolates will be further characterized by performing real-time PCR to determine the presence of any of the following β-lactamase genes: *Klebsiella pneumonia* Carbapenemase gene *bla*KPC, the New Delhi metallo-β-lactamase gene *blaNDM-1*, Verona integron-encoded metallo-β-lactamase gene *bla*VIM, or Oxacillinase-48-like gene *bla*OXA-48-like. In addition each isolate will be screened for the presence of the colistin resistance gene *mcr*-1. The minimum inhibitory concentration (MIC) along with the interpretation for these isolates will be determined using the broth microdilution method.

Collection

Submit a pure culture of reference isolates for testing. Isolates from the *Enterobacteriaceae* family should be resistant to imipenem, meropenem, doripenem or ertapenem by standard susceptibility testing (i.e., minimum inhibitory concentrations of ≥ 4 µg/mL for doripenem, imipenem or meropenem or ≥ 2 µg/mL for ertapenem). *Proteus, Providencia or Morganella spp* should demonstrate resistance to doripenem, meropenem or ertapenem to be considered acceptable for testing as these isolates have an intrinsic resistance to imipenem. These isolates demonstrating only imipenem resistance will be rejected. For *Pseudomonas aeruginosa* isolates the suspect organisms must have an MIC of ≥ 8 µg/mL for doripenem, imipenem or meropenem. Mucoid isolates will be rejected.

Isolates not demonstrating Carbapenem resistance will be rejected unless they have demonstrated Carbapenemase production by a phenotypic test (MHT, mCIM, CarbaNP, etc.).

When submitting an isolate for testing, the organism identification and the AST results should be included on the submission form. Please minimize the number of passages of the suspect isolate as these plasmid based resistance enzymes are easily lost on serial passages.

Shipment of Specimens

Isolates should be transported on an agar plate or slant. Transport specimens to the ASPHL at ambient temperatures. See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

An isolate positive by the modified Carbapenem Inactivation Method (mCIM) indicates the isolate is producing Carbapenemase. PCR testing of the isolate will provide additional information as to the type of the Carbapenemase present. If none of the genes mentioned above are detected by PCR but the isolate is mCIM positive, the sample results suggest the Carbapenemase activity may be the result of a novel mechanism. In such cases the isolate may be forwarded to a reference facility for further characterization.

A report will be sent to the submitting agency and to the Bureau of Epidemiology and Disease Control. Antibiotic susceptibility results will be provided for the isolate to aid in infection prevention measures. These results should not be used as a substitute for diagnostic procedures or used to guide clinical decisions.
Culture Independent Diagnostic Test (CIDTs) Isolate Recovery

For organism recovery from CIDT specimens refer to the Enteric Culture section on pg. 14.

Diphtheria

*Corynebacterium diphtheriae*

Collection

Both throat swabs and nasopharyngeal swabs should be collected from patients suspected of having diphtheria. Wound swabs are also acceptable for cutaneous diphtheria.

The swabs should be placed in semi-solid transport media such as Stuart’s media or Amie’s gel transport media and sent to the ASPHL to be received within 24 hours of collection. The ASPHL must be notified 24 hours in advance (if possible) of a specimen submission.

Shipment of Specimens

The specimen should be transported immediately to the ASPHL or inoculated onto proper media prior to submission.

See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

Cultures will be examined for 48 hours and observed daily for typical growth characteristics. Suspicious colonies are checked for typical morphology and identification of suspected isolates will be made using matrix assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF) and select biochemical tests. Positive cultures of *C. diphtheriae* are reported as presumptive positive and forwarded to the CDC for confirmation and toxigenicity testing. Cultures will be held for at least 48 hours before reporting as negative.

Positive detection of *C. diphtheriae* will be telephoned to the submitting agency and the Bureau of Epidemiology and Disease Control.
Enteric Culture

**Collection**

The most cultured sources for enteric diseases are feces, blood, and urine. Other extra-intestinal sources may be infected with enteric disease organisms. Purulent material from wounds or abscesses may be swabbed or aspirated for the presence of *Salmonella* sp.

- Stool specimens should be taken early in the course of illness when the causative agent is likely to be present in the largest numbers. Freshly passed stool is better than rectal swabs since there is less chance of improper collection and mucus and bloodstained portions can be selected for culture. Collect a small portion of feces, approximately the size of a marble, or a swab coated with feces and place in a transport medium. Whenever possible, multiple specimens should be cultured. The ASPHL will provide agencies with Cary-Blair transport medium. Cary-Blair, or Modified Cary-Blair, is the best overall transport medium for diarrheal stools. Preserved stools (i.e., Cary-Blair or Modified Cary Blair) must be refrigerated during transport and should be received and processed within 3 days of collection. Unpreserved stool is only acceptable if received within two hours of collection.

- Stool specimens previously tested using culture independent tests (CIDTs) will be tested to isolate the organism(s) identified by the submitter's CIDT results. Not all organisms represented in the CIDT results are covered by the ASPHL protocol; contact the ASPHL laboratory for a list of organisms covered by this protocol. CIDT specimens must be submitted in a manner (transport media and timeframe) that is acceptable/appropriate for the organism(s) for which testing is to be conducted (i.e., preserved stools in Carey Blair or Modified Cary Blair refrigerated during transport and received and processed within 3 days of collection). Please include any preliminary results.

**Shipment of Specimens**

Specimens held in transport media should be refrigerated until examined. Transport specimens to the ASPHL at a refrigerated temperature in the proper transport media. Preserved stools should be received and processed within 3 days of collection. Due to the 5 work day operation of the laboratory, receipt of stool specimens on a Friday or the day before a holiday should be avoided if possible to allow for the performance of culture enhancements.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Stool samples, unless otherwise specified, will be screened for *Salmonella, Shigella, Campylobacter, Shiga toxin-producing E. coli*, and upon request *Aeromonas, Pleistomonas, Yersinia*, and *Vibrio*. Cultures are examined daily for 72 hours for characteristic morphology. Suspect colonies are screened biochemically or by matrix assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF), and confirmed with serologic agglutination (where applicable).
Section 1: Bacteriology

**Haemophilus**

**Collection**

*H. influenzae*

Specimens must be collected and cultured as soon as possible since the organisms do not survive well. Pure culture isolates from sterile sites such as blood or cerebrospinal fluid may be submitted to the ASPHL. It is not recommended that clinical materials be submitted to ASPHL for isolation. Clinical samples are referred to the Minnesota Public Health Laboratory for molecular detection and/or serotyping.

Note: Isolates should be transported on chocolate slants.

**Shipment of Specimens**

Reference isolates should be transported on slants of chocolate or Levinthal agar. Both *H. aegyptius* and *H. ducreyi*, because of their fastidious nature, should be transported on chocolate agar slants supplemented with 1% IsoVitaleX.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

*H. influenzae* serotype b (Hib) has been identified as the leading cause of bacterial meningitis and epiglottitis. It has also been implicated as a major cause of pericarditis, pneumonia, septic arthritis, osteomyelitis, and facial cellulitis, as well as an occasional cause of urinary tract infection in children less than 5 years of age. Non-encapsulated strains may cause noninvasive respiratory infections in healthy children, and community acquired pneumonia and chronic bronchitis in adults. Suspect isolates are screened biochemicaly or by matrix assisted laser desorption ionization time of flight mass spectrometry (MALDI-TOF), and confirmed with serologic agglutination (where applicable).

Serotyping is relevant only for the encapsulated strains of *H. influenzae* from sterile sites. Testing is performed for children under 5 years of age using a combination of rapid agglutination techniques in type-specific antisera and/or by PCR. ASPHL will only test for serotype b. All other serotypes will be determined by Minnesota Public Health Laboratory.

Serotype b is called to Bureau of Epidemiology and Disease Control.
Section 1: Bacteriology

**Legionella**

**Collection**

*Legionella* are most frequently isolated from specimens originating in the respiratory tract. On rare occasions, they may be isolated from extra-pulmonary sites including pericardial fluid, peritoneal fluid, wounds, and abscesses. *Legionella* are not known to colonize humans, and therefore are not commensals of the respiratory tract. Please send Legionella isolates from clinical specimens (sputum, transtracheal aspiration, non-respiratory sites, etc.) to ASPHL for identification. Isolates will be forwarded to the CDC for identification and serotyping (if applicable).

**Shipment of Specimens**

Prior notification to the laboratory is requested. Isolates should be shipped on BCYE slants or plates at 4°C on cold packs.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Isolates are forwarded to CDC for identification and serotyping.
Leptospira

ASPHL no longer performs testing for Leptospira species; samples are forwarded to the CDC for testing. Samples for molecular testing may be submitted to the CDC without prior approval; however, culture testing for identification and genotyping does require preapproval from the CDC.

Collection

Blood, cerebrospinal fluid (CSF), and urine are the specimens of choice for recovery of Leptospira. The most appropriate choice is to culture during the first 10 days of illness, and to collect blood and CSF. The specimens should be collected prior to antibiotic treatment and while the patient is febrile. After the first week of illness, the optimal source for isolation of Leptospira is urine.

If culture is requested and culture medium is not available, blood can be collected in tubes containing citrate or EDTA.

Shipment of Specimens

For molecular detection of Leptospira, a minimum of 250 µL of blood (in EDTA or sodium citrate), CSF or serum should be collected. A minimum of 10 ml of urine should be submitted. All samples submitted for molecular testing should be kept frozen at -20 °C.

If culture is desired the specimen (blood, tissue and urine) should be inoculated into Ellinghausen-McCullough-Johnson-Harris (EMJH) semisolid media or Fletchers medium optimally at the bedside but within 48 hours of collection. Specimens should be stored at room temperature and transported at 4°C. Urine should be inoculated within two hours, especially if the urine is acidic. ASPHL does not supply EMJH or Fletchers medium for Leptospira culture.

Isolates submitted for testing should be shipped on Ellinghausen-McCullough-Johnson-Harris (EMJH) at room temperature.

See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

All Leptospira cultures and molecular testing requests are forwarded to CDC.
Section 1: Bacteriology

**Listeria**

**Collection**

Clinical specimens will be accepted only by prior approval of the Bureau of Epidemiology and Disease Control during an outbreak investigation. Clinical specimens from normally sterile sites such as blood, CSF, amniotic fluid, placenta, or fetal tissue do not require special procedures for collection or transport. Specimens from non-sterile sites, such as meconium, feces, and vaginal secretions, respiratory, skin or mucous swabs require prompt handling to prevent the overgrowth of contaminants.

Culture specimens from sterile sites can be plated directly to tryptic soy agar containing 5% sheep, horse, or rabbit blood. Samples for blood culture can be inoculated directly into conventional blood culture broth.

**Shipment of Specimens**

Specimens from sterile sites should be transported as soon as possible. If processing is delayed, specimens should be held at room temperature or at 4 °C for no longer than 48 hours. Specimens from non-sterile sites require prompt handling. If processing is delayed, the materials should be kept at 4 °C or frozen at -20 °C if testing delays are expected to exceed 48 hours. Ship at 4 °C.

Non-sterile specimens (other than stool) can be stored at 4 °C for up to 48 hours. For longer periods of storage, freezing specimens at -20 °C is recommended.

Stools should be shipped overnight at room temperature, if longer delays are expected the sample should be frozen on dry ice.

Reference cultures/isolates can be transported on nutrient agar slants or other non-glucose containing agar at ambient temperature.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Inoculated media will be incubated for 72 hours and examined daily for growth. Isolates and reference specimens are streaked to a blood agar plate and examined daily for typical growth characteristics. Identification of *Listeria* is made based on colony and microscopic morphology, matrix assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF) and/or various biochemical reactions. Once identification is complete the organism is sent to CDC for confirmation and/or serotyping.
**Meningococcus**

*Neisseria meningitidis*

**Collection**

*Neisseria meningitidis* may be isolated from numerous body sites including CSF, blood, petechial aspirates, biopsy samples, joint fluid, and conjunctival swabs. At this time the ASPHL only tests isolates from sterile body site. It is not recommended that clinical materials be submitted to ASPHL for isolation. Clinical samples are referred to the Minnesota Public Health Laboratory for molecular detection and/or serotyping.

Isolates may be submitted on nonselective medium such as chocolate agar, Amie’s, Stuarts or equivalent transport media that is supportive of their growth.

**Shipment of Specimens**

Transport reference isolates as quickly as possible to the ASPHL at room temperature. It is recommended that the containers be insulated during very hot or very cold weather. All cultures must be transported with minimum delay since viability is readily lost. If specimens must be transported from a distant location, the inoculated media must be incubated 18 – 24 hours before transport, and the specimen should arrive within 48 hours.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Cultures are examined daily for typical growth characteristics. Isolates are identified biochemically and/or by MALDI-TOF. Isolates will be forwarded to Minnesota Department of Health for additional characterization or confirmation.

Reports are sent to submitting client and Bureau of Epidemiology and Disease Control.
Section 1: Bacteriology

**Pertussis**

*Bordetella pertussis, B. parapertussis, B. holmesii*

**Collection**

The ASPHL is only offering PCR for the detection of *Bordetella pertussis, Bordetella parapertussis* and *Bordetella holmesii*. The specimen of choice for the recovery of *Bordetella pertussis* and *B. parapertussis* from the respiratory tract is secretions collected from the posterior nasopharynx. Specimens collected from the throat are not acceptable. Specimens collected from the nose are not optimal.

One or two perinasal swab specimens are collected by passing the swabs through the nares as far as possible into the nasopharynx. Leave the swab in place for up to 30 seconds. If resistance is encountered during insertion, try the other nostril. Rotate the swabs for a few seconds, and gently withdraw them.

Use polyester NP swabs – swabs with cotton tips and wooden shafts are not permitted. Specimens collected with swabs made of calcium alginate are not acceptable as this material is inhibitory to the *Bordetella* spp. PCR.

Push the swab, post collection, into a tube of Regan-Lowe semi-solid transport agar. When placing the swab into the tube break/snap/cut the shaft so that it fits properly into the tube. Swab shafts that are force or coiled into the tube may result in the sample being rejected as this creates that potential for laboratory contamination during PCR processing. Leave the swab submerged during transport to the laboratory.

Only patients with signs and symptoms consistent with pertussis should be tested by PCR to confirm the diagnosis. For guidance in distinguishing signs and symptoms of pertussis from those of other conditions, see [http://www.cdc.gov/pertussis/clinical/features.html](http://www.cdc.gov/pertussis/clinical/features.html).

Testing asymptomatic persons should be avoided as it increases the likelihood of obtaining falsely-positive results. Asymptomatic close contacts of confirmed cases should not be tested and testing of contacts should not be used for post-exposure prophylaxis decisions.

When possible, test patients for pertussis during the first 3 weeks of cough, when bacterial DNA is still present in the nasopharynx. After the fourth week of cough, the amount of bacterial DNA rapidly diminishes, increasing the risk of obtaining falsely-negative results by PCR. For more information on diagnostic testing, see [http://www.cdc.gov/pertussis/clinical/diagnostic-testing/diagnosis-confirmation.html](http://www.cdc.gov/pertussis/clinical/diagnostic-testing/diagnosis-confirmation.html).

PCR testing after 5 days of antibiotic use is unlikely to be of benefit; PCR testing following antibiotic therapy can result in falsely-negative findings, although the exact duration of positivity following antibiotic use is not well understood.

**Shipment of Specimens**

Swabs for PCR testing should be collected and transported at 4 °C in Regan-Lowe semi-solid transport agar.
Section 1: Bacteriology

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Identification is made based upon the detection of PCR targets associated with each respective organism (*Bordetella pertussis, Bordetella parapertussis* or *Bordetella holmesii*). Positive PCR results are telephoned to the submitting agency and a notification email is sent to the Bureau of Epidemiology and Disease Control.

**Shigella**

**Collection**

Reference isolates can be submitted for identification/confirmation and/or serogrouping; the submission of reference isolates for archive purposes is no longer necessary.

Refer to the Enteric Culture section for submission of samples where *Shigella* was detected by a culture independent diagnostic test (CIDT).

**Shipment of Specimens**

Reference isolates should be submitted on a nonselective media that is supportive of their growth. Submitted media should be shipped at room temperature.

See section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

*Shigella* isolates are identified biochemically. Isolates biochemically consistent with *Shigella* are serogrouped using type-specific antisera. On occasion isolates may be forwarded to a reference laboratory (e.g., CDC) for additional characterization or confirmation. At this time ASPHL only performs serogrouping; serotyping of isolates is not being performed routinely. Upon the completion of testing the isolate will be reported as *Shigella flexneri, Shigella sonnei, Shigella boydii, Shigella dysenteriae* or as appropriate if another identification is determined (e.g., inactive *Escherichia coli*).
Section 1: Bacteriology

Shiga Toxigenic *E. coli*

**Collection**

Stool specimens should be taken early in the course of illness when the causative agent is likely to be present in the largest numbers. Freshly passed stool is better than rectal swabs, and mucus and bloodstained portions can be selected for culture. Use a small amount of stool or rectal swab to inoculate a MacConkey (MAC) Broth or Gram Negative (GN) Broth and incubate at 35 – 37 °C for 18 – 24 hours before submitting to ASPHL.

Transfer reference isolates of Shiga toxin-producing organisms to a TSA or Nutrient Agar slant and forward to the ASPHL. A GN or MAC broth may be submitted for isolation and confirmation of a toxin-producer. For Shiga toxin-producing E. coli detected from culture independent tests (CIDTs) refer to the Enteric Culture section.

**Shipment of Specimens**

GN or MAC broth may be submitted within seven days of inoculation, if stored and transported refrigerated (2 – 8 °C). Cultures of pure isolates should be submitted within 48 hours of inoculation on nonselective media at room temperature.

Refer to the Enteric Culture section for submission of samples where Shiga toxigenic *E. coli* was detected by a culture independent diagnostic test (CIDT).

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Stool samples, unless otherwise specified, will be screened for *E. coli* O157:H7 and other Shiga toxin-producing organisms. Cultures are examined daily for 72 hours for characteristic morphology. Suspect colonies are screened biochemically, by EIA and/or PCR, and then confirmed with serologic agglutination (where applicable) or forwarded to CDC. Reports will include results of the presence or absence of Shiga-toxin and virulence factors.

For outbreaks, all reports of Shiga toxin-producing organisms are telephoned to the submitting agency and to the Bureau of Epidemiology and Disease Control.
Section 1: Bacteriology

**VISA / VRSA**

*Staphylococcus aureus*

**Collection**

Submit a pure culture of a reference isolates for epidemiological studies or confirmation of Vancomycin-Intermediate/Resistant *Staphylococcus aureus* (VISA/VRSA)

**Shipment of Specimens**

Isolates should be transported on a blood agar plate or slant. Transport specimens to the ASPHL at ambient temperatures. When submitting isolate for testing, the organism identification and the previously determined MIC(s) should be included on the submission form.

See Section 8: Sample Submission Guidelines

**Reporting and Interpretation of Results**

*Staphylococcus aureus* that have developed resistance to methicillin are referred to as MRSA; they are also resistant to most antibiotics commonly used for *Staphylococcus* infections. These drugs include methicillin, oxacillin, nafcillin, cephalosporins, imipenem, and other beta-lactams. The infection is then generally treated with vancomycin. Most isolates of *S. aureus* are susceptible, but use of vancomycin can lead to the development of resistance as well. The minimal inhibitory concentration (MIC) of vancomycin required to inhibit these strains is typically ≤2 micrograms/mL (µg/mL). In contrast, *S. aureus* isolates for which vancomycin MICs are 4-8 µg/mL are classified as vancomycin-intermediate (VISA), and isolates for which vancomycin MICs are ≥16 µg/mL are classified as vancomycin-resistant (VRSA). The definitions for classifying isolates of *S. aureus* are based on the interpretive criteria published in January 2019 (M100 29th Ed.) by the Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS). Possible VRSA isolates with an MIC ≥ 8 µg/mL will be sent to the CDC.

All VISA/VRSA organisms are reported to the submitting agency and to the Bureau of Epidemiology and Disease Control.
Vibrio

Collection

Stool specimens should be collected early, preferably within 24 hours of onset of illness, and before administration of antibiotics. Rectal swabs or fecal material should be placed in the semisolid transport medium of Cary-Blair, or Modified Cary-Blair. Submitted specimens well are treated as an enteric culture, see Enteric Culture section for additional information.

Shipment of Specimens

Specimens in transport media should be shipped to the ASPHL as soon as possible (preserved stool should be submitted within 3 days of collection) at ambient temperature. *Vibrio* isolates may also be submitted for identification; in addition to identification confirmation, *V. cholerae* isolate will also be serotyped.

For submission of samples where *Vibrio* was detected by a culture independent diagnostic test (CIDTs) refer to the Enteric Culture section.

See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

Cultures suspected to contain *V. cholerae*, and other *Vibrio* species are tested with biochemical systems and/or by MALDI-TOF. Cultures presumptively identified as *V. cholerae* will be tested against specific antisera to determine the serogrouping of the isolate. *V. cholerae* strains will fall into two groups based on this serological testing. O group 1 strains (O1) are associated with epidemic cholera; non-O1 strains may cause cholera-like and other illnesses, but are not involved in epidemics. *V. cholerae* O1 strains are divided into three subtypes: Ogawa, Inaba, and Hikojima. The O1 strains are further divided into two biogroups: classical and El Tor. All *V. cholerae* O1 strains are sent to CDC for toxigenic testing.

Positive stool cultures of *V. cholerae* are called to the submitting client and Bureau of Epidemiology and Disease Control.
Section 2: Mycobacteriology

The ASPHL provides diagnostic and reference services for the isolation and identification of *Mycobacterium tuberculosis* (MTB) and other mycobacteria at no charge to all public and private health care providers in the state. The ASPHL receives a federal grant to support the statewide testing of *Mycobacterium* in support of the National Action Plan for the Elimination of Tuberculosis in the United States.

**Collection**

Use a clean, sterile, leak-proof disposable screw-capped 50 mL conical centrifuge tube supplied by the ASPHL. Do not use waxed containers. More detailed information regarding how to obtain specimen collection and submitting materials can be found in Section 8: Sample Submission Guidelines and Section 9: Requesting Collection Kits and Mailing Containers. *Samples will not be processed for Mycobacteria if not received in the laboratory within 7 days of collection.*

**Sputum**

In pulmonary tuberculosis and the related Mycobacterial diseases, sputum (phlegm from deep in the lungs) is the specimen of choice. A 5 - 10 mL sample of sputum is the desired volume for a single examination. Pooled specimens collected over several hours are not suitable for examination. A series of 3 early morning specimens, collected in clean sterile containers, on **consecutive days** should be obtained. Collect the initial specimens before antimicrobial therapy is started. Do not use fixatives or preservatives. Please indicate on the submission form if the sputum has been induced.

**Urine**

The specimen of choice is a clean catch, midstream, first morning specimen. Urine should be collected in a clean, sterile, screw-capped plastic container. Pooled specimens or 24-hour urines are unacceptable. A series of first morning specimens should be collected on three consecutive days.

**Stools**

Stool specimens will be subjected for microscopic analysis only. Reference isolates submitted from stool will be rejected.

**Gastric Washings**

Gastric washings are specimens of last resort because they are highly diluted with gastric fluid, which is damaging to the tubercle bacillus. Specimens should be delivered to the laboratory immediately so neutralization procedures can begin. **These samples are not suitable for mailing.**
Specimens from Sterile Sites

These include cerebrospinal fluid (CSF), pleural fluid, ascitic fluid, joint fluid, pus, exudates, biopsy, and autopsy tissues. These are all surgical specimens and should be collected or taken by a physician or surgeon and placed in sterile containers. Tissue may be delivered in sterile saline. Do not add any preservatives. **Swabs are not optimal for the recovery of Mycobacteria. They are acceptable only if a specimen cannot be collected by other means. A comment will be added to the final report.**

Shipment of Specimens

After collection, identify the specimen with the patient’s name collection date AND collection time. Fill out the proper laboratory submission form, *Microbiology Submission Form* (located at www.azdhs.gov/lab). Include the patient’s name, date of birth, submitting agency, test request, and other pertinent demographic information.

Specimens should be refrigerated immediately after collection, prior to shipment. If specimens are to be shipped, it is necessary to place the specimen in a triple-packed mailing container to avoid contamination in the event of leakage. The desired mailing container consists of a primary screw cap collection tube, a secondary metal screw-capped container placed within a tertiary screw-capped cardboard outer mailer. These containers are provided by the ASPHL upon request. Place the submission form around the outside of the secondary metal container. Never place the form around the primary container. The triple-pack mailer is a safety requirement and a postal shipping mandate. Mail specimens as soon as possible after collection to avoid overgrowth of contaminating bacteria.

Reference specimens may be submitted in tubed solid media or in a liquid culture medium, including Bactec, MGIT, MB-Bacti, and Septi-Chek. Reference specimens that are mailed or delivered by courier transport must be placed in a double mailing container. In the event of courier transportation, the specimen may be sent in a 50 mL conical centrifuge tube inside an inner metal container and then placed in a sealed plastic bag. Securely tighten all caps.

See Section 8: Sample Submission Guidelines

**Reporting and Interpretation of Results**

Specimens are processed daily, five days (Monday – Friday) a week. Smears are examined daily by fluorescent microscopy, using a fluorochrome stain. The results of positive smears on all new patients are telephoned to the submitting agency within 24 hours from receipt with the exception of specimens received on Friday afternoons or on an afternoon prior to a holiday. Preliminary laboratory reports are prepared and sent out for all smear results.

Specimens are cultured onto both solid and liquid media. Cultures are examined for growth during a period of six weeks (on negative smears) and eight weeks for positive smears, before being reported as “No Mycobacteria isolated”. Cultures exhibiting typical colonial morphology are identified using matrix assisted laser desorption/ionization – time of flight mass spectrometry (MALDI-TOF). Real time PCR/melt curve analysis can be performed on cultures from liquid media (MGIT broths) for the detection of MTBC. Allow 48 hours after detection of growth for identification of the organism. Nucleic Acid Amplification (Cepheid GeneXpert MTB/RIF Molecular Assay) test is automatically
performed on smear positive **sputum** specimens from new patients on samples processed at the ASPHL. The Cepheid GeneXpert MTB/RIF Molecular assay may also be performed on smear negative **sputum** specimens from new patients upon approval from the Arizona Department of Health Services, TB control section main telephone line at (602)-364-4750. Submitters who process their own samples may also submit samples to the ASPHL for a Nucleic Acid Amplification (NAA) assay with the approval of the TB Control Section. Once approval has been given, the submitter must send a fresh sample to be processed by the ASPHL. Testing can only be performed on samples processed by N-acetyl L- cysteine sodium hydroxide (NALC-NaOH) method. Performance characteristics using samples processed by other means (ex. Oxalic acid, cetylpyridinium chloride, etc.) have not been established in the laboratory and therefore cannot be tested by the GeneXpert. In addition, performance of the test has not been evaluated on pediatric patients. Positive NAA test results are telephoned to the submitting agency within 48 hours from receipt with the exception of specimens received on Friday afternoons or on an afternoon prior to a holiday.

**Drug Susceptibilities**

Drug susceptibilities are performed only on *Mycobacterium tuberculosis complex* (MTBC). If the MTBC is resistant to any of the first-line drugs tested by MGIT DST, an indirect susceptibility is performed by the conventional agar proportion method, where an additional drug regimen is tested. Drugs tested by the MGIT method are isoniazid, rifampin, ethambutol and pyrazinamide. These results are reported within 17 days of MTB identification from culture for all initial diagnostic specimens. The agar proportion method includes the same drugs plus ethionamide, ofloxacin, and capreomycin. Taken together, final results for susceptibility is between three to six weeks from identification. Susceptibilities are performed every three months on specimens that remain positive for MTBC. MGIT susceptibility results are telephoned to the submitter. Susceptibility testing should be requested on subsequent isolates when a regimen appears to be failing. The manifestations of a failing regimen are: lack of conversion of smear and culture to negative within three months for persons receiving regimens containing both isoniazid and rifampin; lack of conversion of smear and culture to negative after five months for those receiving other regimens (without both isoniazid and rifampin); smears and cultures showing a decrease in number of organisms or colonies followed by a persistent increase in numbers (“fail and rise”).

**Note:** When submitting MGIT samples for MTB drug susceptibility testing, indicate the date the MGIT instrument identified the sample as positive. Omitting this information will cause delays in susceptibility testing.

Isolates of MTBC and NAAT positive processed samples will be submitted to the CDC for molecular detection of drug resistance (MDDR) service if one of the following criteria is met:

1. By patient history if there is a high-risk of rifampin resistance or multidrug resistance MTBC
2. Known rifampin resistance (by rapid test or culture-based DST)
3. Patients where the result of drug resistance will predictably have a high public health impact (e.g., daycare workers, nurses)
4. Patient is known to have certain adverse reactions to critical anti-TB drug (e.g., allergy to rifampin)
5. Mixed or non-viable cultures
6. Isolates which fail to grow in DST medium
Section 2: Mycobacteriology

7. Other situations considered on case by case basis

The results of all specimens are reported to the submitter, including results from the CDC. In addition, all positive results and susceptibility are reported to the Tuberculosis Elimination Section of the Bureau of Epidemiology and Disease Control, Arizona Department of Health Services.
Section 3: Limited Parasitology

Intestinal and blood parasites are diagnosed mainly by morphologic examination of diagnostic stages of the microorganism. Properly collected and preserved specimens are of the utmost importance, since old or poorly preserved materials are of little value in establishing a diagnosis and may lead to erroneous conclusions. The ASPHL no longer accepts routine diagnostic samples. The laboratory offers screening for *Giardia* and *Cryptosporidium* to assist in outbreak investigations with approval from the Bureau of Epidemiology and Disease Control. All other submissions are forwarded to CDC with the approval of the Bureau of Epidemiology and Control.

For testing assistance for organisms/diseases not offered by the ASPHL, please refer to the Centers for Disease Control and Prevention (CDC) Test Directory website [http://www.cdc.gov/laboratory/specimen-submission/list.html](http://www.cdc.gov/laboratory/specimen-submission/list.html). Information regarding available tests, appropriate specimen type(s), collection and storage conditions, shipping requirements and appropriate test authorizations can be found on this website. Specimens should be forwarded to the CDC from the ASPHL and should not be sent directly from other facilities; please contact the Bacteriology section with any questions or for sample coordination with bacteriology or parasitology related samples.

**Collection**

**Fecal specimens (***Giardia* and *Cryptosporidium**)**

Collect the stool in a clean container or on clean paper, and then transfer to transport in Cary-Blair. Follow the instructions included with the containers. Mix thoroughly to assure adequate fixation. Do not contaminate specimen with urine or dirt. Administration of barium, magnesia, or oil before collection will render the specimen unsuitable for testing. Label each vial with patient’s name and address. Because the host passes parasites intermittently, multiple specimens should be examined. These irregularities emphasize the need to collect at least three specimens over 10 to 14 days.

**Blood Parasites**

Blood smears are best made from blood not containing anticoagulants, since anticoagulants can interfere with parasite morphology and staining. For routine diagnosis, a thick film is preferable; however, parasite morphology is more distinct and typical when observed in a thin film. Therefore, it is important to collect both thick and thin films for submission. Thin films are made by depositing a single drop of blood at one end of the slide and spreading it across the slide in preparation for a differential count. Thick films are prepared by touching the under-surface of a slide with a fresh drop of blood from a finger (without touching the skin) and rotating the slide to form a film about the size of a dime. Alternately, several drops of blood can be deposited at the end of a slide and puddle with an applicator stick or toothpick. Allow 8 – 12 hours drying time for a thick film before staining. Giemsa Stained slides should be placed in a cardboard slide holder, and labeled with proper identification.
Section 3: Limited Parasitology

If necessary, thick and thin smears can be prepared from anticoagulated blood, but the staining characteristics are not as good. EDTA anticoagulated blood is better for staining than citrate or heparin anticoagulant.

The time of specimen collection is important with malaria, but less important in other filarial infections. Malaria parasites are most numerous about midway between chills. One specimen taken at this time and a second specimen collected 5 – 6 hours later are ideal. Because of nocturnal periodicity in filarial infections, the specimen should be taken between 10 PM and 2 AM. In *Loa loa*, there is diurnal periodicity, and these specimens should be collected between 10 AM and 2 PM.

**Free-Living Amoebae**

Unfixed specimens for culture (fresh, unfixed tissue and paraffin-embedded and formalin-fixed tissue, cerebrospinal fluid (CSF), biopsy specimen, deep corneal scrapings, and ocular fluids) should be sent overnight at ambient temperature. Unfixed deep scraping and biopsy materials for identification of free-living amoeba are usually very small and may dry if they are not stored in proper fluid such as 0.5x PBS or “amoeba saline”. These specimens should be transported to the laboratory within 24 hours. Fixed specimens, including those in 70-90% ethanol, should be sent overnight on ice packs. Culture plates and slides must be packaged carefully to prevent breakage. All samples are forwarded to CDC for identification and/or confirmation. For additional information on available test, specimen collection and storage, as well as shipping requirements refer to the CDC Test Directory website (http://www.cdc.gov/laboratory/specimen-submission/list.html).

**Shipment of Specimens**

Fill out the *Microbiology Submission Form* located at: [http://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving](http://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving)

Include the patient’s name, date of birth, address, submitting agency, test request, and other pertinent information on the form. Identify the specimen with the patient’s name and date of collection. Make sure that identification on the specimen matches the form. **Include travel history with the request for blood parasite identification.**

Specimens sent through the mail must be in containers that meet postal regulations for infectious materials. Specimen containers should be placed inside a double mailing container, which consists of an inner metal case with a screw cap placed within a screw-capped outer cardboard container.

Mailed stool specimens require use of one vial containing 10% formalin and a two-vial method of collection and shipping is advocated. **If you are submitting a stained slide for identification or confirmation, please include the original preserved sample for additional testing.**

See Section 8: Sample Submission Guidelines
Section 3: Limited Parasitology

**Reporting and Interpretation of Results**

Specimens received for parasitology other than *Giardia* and *Cryptosporidium* will be forwarded to CDC with the approval of the Bureau of Epidemiology and Disease Control. Blood smears will be forwarded on to the CDC for confirmation of results. A preliminary report will be generated by the ASPHL indicating that the specimen has been forwarded on to the CDC. The final report will be generated upon issuance of a report from the CDC.
Section 4: Serology

The Serology Section is responsible for performing diagnostic testing for communicable diseases in support of outbreak investigations, and reference testing for private and public laboratories. The time required to process a serology specimen varies considerably, as indicated by the following table. Detailed information on the collection and submission of laboratory samples for any of the following tests can be obtained in the narrative guidelines that follow.

<table>
<thead>
<tr>
<th>Organism/Disease</th>
<th>Specimen Type</th>
<th>Minimum Sample Volume</th>
<th>Test Method</th>
<th>Comments</th>
<th>Turn Around Time (TAT) Business Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dengue virus*1,5</td>
<td>Serum</td>
<td>1.0 mL</td>
<td>IgM EIA, PCR, PRNT</td>
<td>Date of onset needed, test method determined by case history. PRNT testing performed on presumptive positive or equivocal samples. Specimen may also be tested for West Nile virus and St. Louis Encephalitis virus.</td>
<td>2-7 days, 2-7 days, 14-28 days</td>
</tr>
<tr>
<td>Chikungunya virus*5</td>
<td>Serum</td>
<td>1.0 mL</td>
<td>IgM EIA, PCR, PRNT</td>
<td>Date of onset needed, test method determined by case history. PRNT testing performed on presumptive positive or equivocal samples.</td>
<td>2-7 days, 2-7 days, 14-28 days</td>
</tr>
</tbody>
</table>
## Serology

<table>
<thead>
<tr>
<th>Organism/Disease</th>
<th>Specimen Type</th>
<th>Minimum Sample Volume</th>
<th>Test Method</th>
<th>Comments</th>
<th>Turn Around Time (TAT) Business Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hantavirus*&lt;sup&gt;2&lt;/sup&gt; Serum</td>
<td>Serum</td>
<td>1.0 mL</td>
<td>IgM EIA</td>
<td></td>
<td>1-7 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>IgG EIA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HIV</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>Ag/Ab EIA</td>
<td>ADHS HIV Prevention Program only.</td>
<td>3-5 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>HIV-1, HIV-2 Ab Differentiation</td>
<td>Referred to New York Health Department.</td>
<td></td>
</tr>
<tr>
<td>Measles</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>IgM EIA</td>
<td>Referred to CDC</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>IgG EIA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mumps</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>IgM EIA</td>
<td>Referred to CDC</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>IgG EIA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Q Fever (&lt;i&gt;Coxiella&lt;/i&gt;), phase I &amp; phase II&lt;sup&gt;3&lt;/sup&gt;</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>IgG IFA</td>
<td>Referred to CDC</td>
<td></td>
</tr>
<tr>
<td>Rickettsia spp.&lt;sup&gt;3&lt;/sup&gt;</td>
<td>Paired Serum</td>
<td>0.5 mL</td>
<td>IgG IFA</td>
<td>Acute and convalescent serum required for testing.</td>
<td>Paired sera 2-14 days</td>
</tr>
<tr>
<td>Rocky Mountain Spotted Fever (RMSF) Group</td>
<td>Whole Blood</td>
<td>2.0 mL</td>
<td>PCR</td>
<td>Single sera testing requires prior approval.</td>
<td>Single sera 14-28 days PCR</td>
</tr>
<tr>
<td>Typhus Fever Group</td>
<td></td>
<td></td>
<td></td>
<td>Typhus Fever Group and other spp. referred to CDC.</td>
<td>2-7 days</td>
</tr>
<tr>
<td>Other spp.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rubella</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>IgM EIA</td>
<td>Referred to CDC</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>IgG EIA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>St. Louis Encephalitis (SLE) virus&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Serum</td>
<td>0.5 mL</td>
<td>IgM EIA</td>
<td>Sample will also be tested for WNV</td>
<td>3-7 days</td>
</tr>
<tr>
<td></td>
<td>CSF</td>
<td>0.5 mL</td>
<td>PRNT</td>
<td>PRNT testing is</td>
<td>14-28 days</td>
</tr>
<tr>
<td>Organism/Disease</td>
<td>Specimen Type</td>
<td>Minimum Sample Volume</td>
<td>Test Method</td>
<td>Comments</td>
<td>Turn Around Time (TAT) Business Days</td>
</tr>
<tr>
<td>------------------</td>
<td>---------------</td>
<td>-----------------------</td>
<td>-------------</td>
<td>----------</td>
<td>-------------------------------------</td>
</tr>
<tr>
<td>West Nile Virus (WNV)¹</td>
<td>Serum CSF</td>
<td>0.5 mL 0.5 mL</td>
<td>IgM EIA PRNT</td>
<td>Sample will also be tested for SLE</td>
<td>3-7 days 14-28 days</td>
</tr>
<tr>
<td>Zika virus*¹, ⁴</td>
<td>Serum CSF Whole Blood</td>
<td>2.0 mL 1.0 mL 1.0 mL</td>
<td>IgM EIA PCR PRNT</td>
<td>Testing determined by case history and/or travel history and follows the current guidelines from the CDC PRNT testing performed on presumptive positive or equivocal samples</td>
<td>2-7 days 2-7 days 14-28 days</td>
</tr>
</tbody>
</table>

* Prior notification and approval required

Test abbreviations:
- EIA – Enzyme Immunoassay
- IFA – Indirect Fluorescent Antibody
- PCR – Polymerase Chain Reaction
- Ab – Antibody
- Ag – Antigen
- PRNT – Plaque Reduction Neutralization Test
1. Significant cross-reactivity has been observed within the viruses in the Flavivirus group including Dengue, SLE, WNV, and Zika. Confirmatory testing with Plaque Reduction Neutralization Testing (PRNT) will be performed on samples where cross-reactivity is suspected.

2. Specimens submitted for Hantavirus testing are tested for both IgG and IgM antibodies. Demonstration of the presence of IgM antibody is suggestive of recent exposure to Hantavirus (Sin Nombre Virus). With prior notice and approval from the Arizona Department of Health Services Office of Infectious Diseases, the turnaround time for test results can be shortened.

3. Testing for Spotted Fever group rickettsia, Typhus Fever group rickettsia, or Q fever will only be performed on paired sera. Single serum specimens may be tested but will require approval by the Arizona Department of Health Services Office of Infectious Disease. Serum samples that cannot be tested or shipped to the ASPHL within 2-5 days of collection must be stored at ≤20 °C in a non-self-defrosting freezer. Serum must be removed from the blood clot prior to freezing. Do not freeze blood specimens. With prior approval from the Serology department, acute serum samples can be stored at the ASPHL. Testing will proceed when the convalescent serum sample is received.

4. For Zika virus testing to proceed, serum submission is a requirement. Other specimen types such as urine, whole blood, CSF, and amniotic fluid can also be submitted and recommended but they must be accompanied by a serum specimen. Test method is determined based on epidemiological and clinical information on each patient submitted and the ASPHL follows the most current recommendations from the CDC for Zika virus testing. Approval from local and state health department is required for Zika testing.

5. For Dengue virus and Chikungunya virus testing, patient history including travel, symptoms, and symptom onset is required. The test method utilized for detection of these viruses is dependent on clinical information.

**Specimen Collection:**

**Blood**

Blood specimens should be collected aseptically in an appropriate collection tube and labeled with a patient identifier (e.g., patient name). Follow the manufacturer’s instructions for volume to collect for each tube submitted. For pediatric patients, smaller volumes of blood may be collected utilizing pediatric tubes.

- **Serum:** red top, tiger top, gold top vacutainer tubes
- **Whole Blood/plasma:** lavender top vacutainer tube w/ EDTA anticoagulant

Acute blood samples are should be drawn as soon as possible after appearance of symptoms. A convalescent sample should be drawn 10 – 14 days after the acute sample.

After collection, the tube may be transported directly to the Arizona State Public Health Laboratory (ASPHL) or the tube may be centrifuged and the serum/plasma poured off into a separate vial.
Whole blood specimens for PCR testing should not be centrifuged prior to submission to the ASPHL.

Other

Other specimens may be sent to the ASPHL for serological testing. These include cerebrospinal fluid (CSF) and amniotic fluid which should be collected in a sterile container.

Transportation & Storage:

Store samples refrigerated and ship on cold packs or wet ice. Do not freeze whole blood specimens. The specimen should be transported to the ASPHL as soon as possible. Due to the intense heat observed in the summertime, it is advisable to ship the specimen cold to prevent damage to the specimen during transit.

Samples must be transported with the appropriate paperwork, verifying that the information appearing on the specimen matches that on the submission form. Since the integrity of the sample must be maintained from the time of collection of the sample until testing is completed, labeling errors will result in rejection of the specimen.

Laboratory submission forms should be filled out completely with all pertinent demographic information. Successful tracking of positive cases is reliant on complete and accurate information being supplied on these forms, including patient name or identifier, date collected, date of onset of illness, submitter’s name and address, and agency code.

For HIV serological testing, specimens are to be submitted with an HIV Submission Form only. All other serological specimens should be accompanied with a Microbiology Submission Form.

Specimens may be mailed or delivered by courier to the ASPHL. See Section 8: Sample Submission Guidelines.

If Sent by Courier

- Blood and blood products sent in vacutainer tubes should first be placed in a primary screw-cap leak proof container (such as a 50 mL plastic conical tube available from the ASPHL) to reduce the risk of shattering while in transit.

- The specimen should then be placed in a secondary container such as a plastic specimen bag with separate compartments for the submission form and specimen.

- All infectious material must be triple packaged and conform to U.S. Department of Transportation (DOT) requirements.

- Pack the specimen and its form in absorbent material to help prevent breakage.

  Note: It is acceptable to send more than one specimen together, as long as they are properly secured and identified.
Section 4: Serology

If Sent by Mail

- Blood sent in vacutainer tubes should first be placed in a leak proof primary container (such as a 50 mL conical tube available from the ASPHL) to reduce the risk of shattering while in transit.

- All infectious material must be triple-packaged and conform to current shipping regulations. Consult the Domestic Mail Manual published by the US Post Office (USPS) for current USPS requirements, and the Hazardous Material Regulations (HMR) for current DOT requirements.

- Wrap the submission form around the secondary container, and place inside the tertiary container or cardboard mailer. Package the specimen with enough absorbent material for entire contents and to help prevent breakage.

  **Note:** Do not put the submittal form around the primary container; it must be around the secondary container.

- Place appropriate biohazard label on the outside of the secondary container before transportation to the ASPHL.

50 mL conical tubes and cardboard mailers are available from the ASPHL Receiving Section via request for materials form available at [http://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving](http://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving) or by emailing labreceiving@azdhs.gov. Please submit your orders in advance to ensure prompt service and delivery.

**Rejection Criteria:**

Samples may be considered unacceptable if they are:

- not properly identified or are improperly identified
- grossly hemolyzed (blood cells are lysed)
- contaminated with bacteria
- lipemic
- leaked in transit or broke in transit
- prior approval was not given
- no convalescent serum received.

The submitter will be notified of all rejected laboratory specimens by telephone and with a laboratory report mailed to the submitting agency confirming the reason for rejection.
Section 5: Virology

The Virology Section is responsible for performing diagnostic, reference, and surveillance testing for viruses. The following table provides the viruses the ASPHL Virology department can identify and the turnaround times to report results. Detailed information on the collection and submission of laboratory samples for any of the following tests can be obtained in the narrative guidelines that follow.

Note: bacteriological collection swabs and transport medium are not acceptable for virus detection. Swabs should be placed into a liquid media such as viral transport media or universal transport media.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Specimen</th>
<th>Transport Medium &amp; Volume</th>
<th>Comments</th>
<th>Turn Around Time (TAT) Business Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arbovirus Surveillance</td>
<td>Mosquito Pools Culex spp. Aedes spp.</td>
<td>None</td>
<td>Ship frozen</td>
<td>7-14 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterovirus</td>
<td>Respiratory Swabs¹, Rectal Swab, Viral Isolate</td>
<td>Hanks, Viral Transport Media (VTM), Universal Transport Media (UTM), Sterile Saline. Sterile collection container</td>
<td>Enterovirus D68 is referred to CDC. Must have prior approval.</td>
<td>Referred to CDC</td>
</tr>
<tr>
<td></td>
<td>Stool, CSF</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Influenza virus</td>
<td>Respiratory Swabs¹, Viral Isolates</td>
<td>Hanks, Viral Transport Media (VTM), Universal Transport Media (UTM), Sterile Saline. Sterile collection container</td>
<td>Surveillance only specimens—aliquot of sample can be submitted, minimum volume 0.5 mL</td>
<td>PCR: 1-5 days</td>
</tr>
<tr>
<td>• Seasonal</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Avian</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Novel</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Variant</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>• A only</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

¹ Swabs should be placed into a liquid media such as viral transport media or universal transport media.
<table>
<thead>
<tr>
<th>Organism</th>
<th>Specimen</th>
<th>Transport Medium &amp; Volume</th>
<th>Comments</th>
<th>Turn Around Time (TAT) Business Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Measles virus</td>
<td>Respiratory Swabs¹, viral isolates</td>
<td>Hanks, Viral Transport Media (VTM), Universal Transport Media (UTM), Sterile Saline.</td>
<td>Submission of both a throat swab and urine is preferred for testing.</td>
<td>PCR: 1-5 days</td>
</tr>
<tr>
<td></td>
<td>Respiratory aspirates, urine</td>
<td>Sterile collection container</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Min. volume: 2 mL</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 mL – Urine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mumps virus</td>
<td>Respiratory Swabs¹, viral isolates</td>
<td>Hanks, Viral Transport Media (VTM), Universal Transport Media (UTM), Sterile Saline.</td>
<td>Submission of both a buccal swab and urine is preferred for testing.</td>
<td>PCR: 1-5 days</td>
</tr>
<tr>
<td></td>
<td>Respiratory aspirates, urine</td>
<td>Sterile collection container</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Min. volume: 2 mL</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 mL – Urine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Norovirus PCR</td>
<td>Stool (raw)</td>
<td>Sterile collection container</td>
<td>Do not freeze.</td>
<td>2-10 days</td>
</tr>
<tr>
<td>Rabies</td>
<td>Small animal (bat), animal head, brain tissue</td>
<td>None</td>
<td>Refer to page 44</td>
<td>1-2 days</td>
</tr>
<tr>
<td>Respiratory Virus Panel (PCR)</td>
<td>NP Swab</td>
<td>Viral Transport Media (VTM), Universal Transport Media (UTM)</td>
<td>Prior Approval Required</td>
<td>1-5 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Min. volume: 2 mL</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Organism | Specimen | Transport Medium & Volume | Comments | Turn Around Time (TAT) Business Days
--- | --- | --- | --- | ---
Zika virus³ | Amniotic Fluid Urine | Sterile collection container
Min. volume: 2 mL. | Testing determined by case history and/or travel history and follows the current guidelines from the CDC. | 2-7 days

1. Respiratory Swabs: Nasopharyngeal, throat, buccal
2. Enterovirus D68 requests need prior approval, specimen referred to the CDC.
3. For Zika virus testing, serum must be submitted in addition to other specimen types.

### Collection

In order to optimize the ability of the Virology Section to identify viral agents from clinical specimens, it is very important that the specimens be collected, handled and transported in a manner that minimizes deleterious effects on any viral agents present. In addition, sufficient information should be provided with a submitted specimen to guide the laboratory in the selection of proper inoculation techniques for the viral agents suspected.

#### Transport Media

Transport media for viral detection should be a liquid medium free from serum. The following liquid media is approved for use for viral sample collections

- Hank’s Balanced Salt Solution
- Viral Transport Media (VTM)
- Universal Transport Media (UTM)
- Sterile Saline (only when other media is not available)
Collection Swabs

Recovery of virus is also dependent upon the swab the specimen was collected. Acceptable swabs include:

- Polyester
- Synthetic
- Flocked swabs (FLOQ)
- Dacron
- Nylon

Unacceptable swab types include:

- Calcium alginate swabs
- Cotton tip swabs with wooden shafts

Nasopharyngeal/Throat

Virus isolation is most successful if respiratory specimens are collected within 3 to 5 days of onset of illness. Swabs from both the throat and nasal passage should be collected. The pharynx is swabbed vigorously with a swab moistened with collection medium and then placed in a transport container containing transport medium. Break off the ends of the applicator sticks leaving the swab tips in the collection medium.

NP swabs are used to collect specimens from the nasal passage. Allow the swabs to remain in the nasal passages for a few seconds to absorb the nasal secretions laden with virus. Place the swabs in the Hanks or VTM and label vial.

Store specimens frozen at -70 °C if they cannot be tested within 72 hours. Transport to the ASPHL on wet ice. Do not freeze at -20 °C.
Section 5: Virology

Urine

Urine specimens can be submitted for further aid in diagnosis of a viral infection. Generally Measles, Mumps, and Zika virus can be found in the urine. For measles virus and mumps virus testing, it is recommended that a respiratory specimen and a urine specimen be submitted for a suspect patient.

Collect the specimen as soon as possible after onset of illness. Clean voided specimens (10-20 mL) are collected in sterile containers and transported immediately to the laboratory on wet ice or cold packs.

Throat Washings

Throat washings should be collected by gargling with Hanks Balanced Salt Solution (HBSS). Collect the specimen in a sterile container. Collect the specimen as soon as possible after onset of illness. Transport specimen immediately to the laboratory on wet ice or cold packs.

Cerebrospinal Fluid (CSF)

For virus isolation, 3-4 mL of CSF should be collected no later than 7-10 days after onset of illness. Place in a sterile screw capped tube without collection medium. If delays in transport, store frozen at -70 °C. Transport to the laboratory on wet ice or cold pack.

Stools

Place three to four grams of stool into a sterile container and transport to the laboratory on wet ice or a cold pack.

Stool specimens collected to test for the presence of Norovirus must be refrigerated (not frozen) as soon as possible after collection.

Autopsy or Biopsy Specimens

Autopsy specimens should be collected within 24 hours after death. Samples from probable sites of pathology are collected using separate, sterile instruments and separate sterile containers for each specimen. Tissues are transported to the laboratory on wet ice or cold pack. If they cannot be tested within 48 hours, they should be stored frozen at -70 °C.

Shipment of Specimens

All infectious material must be triple-packaged. Place specimens in screw-cap leak proof primary container. Place primary container in a leak proof secondary container and wrap submission form around secondary container. Place secondary container and submission form in an appropriate tertiary container. Ensure adequate ice or cool packs are used if required. Each specimen must be accompanied with a Microbiology Submission Form. Mail, ship or courier specimens to the ASPHL.

See Section 8: Sample Submission Guidelines.
**Reporting and Interpretation of Results**

Influenza diagnostic samples must be shipped at -70 °C if testing cannot be performed within 72 hours. A disclaimer will be added to all samples submitting for influenza testing that states: “Specimens should be shipped at -70 °C if testing cannot be performed with 72 hours of collection. If shipping conditions are not met, a negative test result does not rule out the presence of Influenza virus.”
Rabies

Collection

Updated collection information can be found at http://www.azdhs.gov/preparedness/epidemiology-disease-control/rabies/index.php

Prior approval from the local health department and Bureau of Epidemiology and Disease Control Vector-Borne and Zoonotic Disease Section is needed on all submissions. Refer to the link above for information on approval.

To identify animals that are rabid, testing requires samplings of brain tissue. For animals that are extremely small such as bats, mice, rats, and other rodents, the animal should be sent intact. Larger animals but no larger than the size of a dog should have the head of the animal submitted. The head should be severed close to the shoulders allowing sufficient tissue of the throat to remain, to ensure inclusion of salivary glands. For larger animals, such as cows, horses, and horned animals, the brain should be removed by a veterinarian and sent to the laboratory. Arrangements can be made with ADHS for removal of brain tissue at the Arizona Veterinary Diagnostic Laboratory in Tucson.

- **Entire Animal:** Bats, mice, rats, rodents
- **Head only:** animals the size of skunks, foxes, wild cats, domestic cats, domestic dogs, javelina, etc.
- **Brain only:** horses, cows, horned animals

**Please Note:** Rodents will be tested only by prior approval from the Vector-Borne and Zoonotic Disease Section of the Bureau of Epidemiology and Disease Control. Rodents may carry other serious and deadly diseases, such as plague, tularemia, or Hantavirus, and should be handled with extreme caution.

Birds and reptiles **will not** be accepted for examination.

Specimens for rabies examination should be collected immediately after the death of the animal. Decomposed specimens or specimens infested with maggots may not be testable but will be determined by the Virology Section.

**Shipment of Specimens**

All infectious material including specimens submitted for rabies testing should be triple-packaged. Place the head in a leak proof primary container, and place the primary container in another leak proof secondary container. The secondary container should be placed in a tertiary container that is filled with wet ice or cold packs as necessary. An animal ID should be written on the primary, secondary, or tertiary container to ID each sample submitted. An approved Rabies Submission Form should be placed in a separate sealed plastic bag outside of the secondary container, or in a separate plastic bag or envelope taped to the outside of the box. Ship the specimens to the Arizona State Public Health Laboratory in Phoenix. Testing delays may be experienced on specimens that are received frozen.

See Section 8: Sample Submission Guidelines.
Section 5: Virology

**Reporting and Interpretation of Results**

In all cases when exposure of a human is reported by a physician or veterinarian, laboratory examination will be made consisting of microscopic examination of smears prepared from brain material. The results of the microscopic examinations will be available 24 to 48 hours after receipt of the specimen. Positive results will be reported by telephone to the Vector-Borne and Zoonotic Disease Section of the Bureau of Epidemiology and Disease Control.
Section 6: Foodborne Pathogens

The Foodborne Pathogens section conducts microbiological examinations of clinical and food specimens for isolation and identification of enteric pathogens of public health significance. Test requests for foodborne viruses and toxins which are not performed in the laboratory will be forwarded to CDC, FDA or FSIS laboratory after coordinating the issue with Bureau of Epidemiology and Disease Control and appropriate federal agency. Sanitarians and representatives of federal, state, county and city agencies responsible for monitoring quality and enforcing regulations governing production and handling of food and water may submit samples for analysis.

Clinical Testing

Upon receipt at the ASPHL, all specimens are logged in and assigned to the appropriate area for processing. The time required to process a microbiology specimen varies considerably, as indicated in Section 1 – Bacteriology table (pgs. 9-10). Detailed information on the collection and submission of laboratory samples on any of the following tests can be obtained in the following narrative guidelines.

During outbreaks, the Bureau of Epidemiology and Disease Control may conduct surveillance to determine extent of the outbreak or to determine the relatedness of microorganisms identified in the outbreak. The Office of Public Health Microbiology will support these outbreak investigations through the use of various tools. Data may be shared in these investigations with other states and the CDC in the event of a multi-state outbreak.

Salmonella

Collection

Submit reference isolates of \textit{Salmonella} for epidemiological studies. Refer to the Enteric Culture section for submission of samples where Salmonella was detected by a culture independent diagnostic test (CIDT).

Shipment of Specimens

Reference isolates should be submitted on a nonselective media that is supportive of their growth. Submitted media should be shipped at room temperature. See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

\textit{Salmonella} isolates are identified biochemically and/or by MALDI-TOF. Isolates are serotyped for epidemiological purposes using whole genome sequencing. On occasion isolates may be forwarded to the CDC for additional characterization or confirmation. The Bureau of Epidemiology and Disease Control is notified upon the detection of Salmonella Typhi.
**Section 6: Foodborne Pathogens**

**Shiga Toxigenic E. coli**

**Collection**

Stool specimens should be taken early in the course of illness when the causative agent is likely to be present in the largest numbers. Freshly passed stool is better than rectal swabs, and mucus and bloodstained portions can be selected for culture. Use a small amount of stool or rectal swab to inoculate a MacConkey (MAC) Broth or Gram Negative (GN) Broth and incubate at 35-37 °C for 18-24 hours before submitting to ASPHL.

Transfer reference isolates of Shiga toxin-producing organisms to a Trypticase Soy Agar or Nutrient Agar slant and forward to the ASPHL. A GN or MAC broth may be submitted for isolation and confirmation of a toxin-producer. For Shiga toxin-producing *E.coli* detected from culture independent tests (CIDTs) refer to the Enteric Culture section.

**Shipment of Specimens**

GN or MAC broth may be submitted within seven days of inoculation, if stored and transported refrigerated (2-8 °C). Cultures or pure isolates should be submitted within 48 hours of inoculation on nonselective media at room temperature.

Refer to the Enteric Culture section for submission of samples where Shiga toxigenic *E.coli* was detected by a culture independent diagnostic test (CIDT).

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Stools samples, unless otherwise specified, will be screened for *E.coli* O157:H7 and other Shiga toxin-producing organisms. Cultures are examined daily for 72 hours for characteristic morphology. Suspect colonies are screened biochemically, by EIA and/or PCR, and then confirmed with serologic agglutination (where applicable) or forwarded to CDC. Reports will include results of the presence or absence of Shiga-toxin and virulence factors.

For outbreaks, all reports of Shiga toxin-producing organisms are reported to the Bureau of Epidemiology and Disease Control and to the submitting agency.
Section 6: Foodborne Pathogens

Food Product Samples

In order to ensure rapid and efficient service, communication with the Foodborne Pathogens Section is very important. Before submitting or shipping any samples for analysis, please call the Arizona State Public Health Laboratory (ASPHL) Foodborne Pathogens Section at (602) 542-6130.

A three-day food history and investigation observation should be used to guide the selection of appropriate foods for analysis. An investigation should be conducted before submitting samples to the lab for analysis.

Collection

After determining the appropriate food specimen to submit, aseptically collect approximately 200 grams of a solid product or about 100 mL of a liquid. Collection should be in a sterile whirl-pak plastic bag or sterile urine collection cup. The ASPHL does not provide sterile collection containers for food collection.

Shipment of Specimens

All samples must be kept cold (<10 °C) during transit to the laboratory. Samples that are shipped should be placed in a leak-proof shipping container, preferably a Styrofoam container, packed with sealed cold packs (e.g. blue ice packs). Samples that are hand delivered on wet ice should be protected from cross contamination as the ice melts during transit.

See Section 8: Sample Submission Guidelines.

A properly completed Food Analysis Submittal Form must accompany each individual sample. [Link](http://www.azdhs.gov/documents/preparedness/state-laboratory/public-health-microbiology/food-analysis-form.pdf)

Each sample must be identified by a unique number that corresponds to the identification number written on the submission form. More detailed information regarding how to obtain collection/submission supplies can be found in Section 9: Requesting Collection Kits and Mailing Containers.

Reporting and Interpretation of Results

Quality control samples are tested for aerobic plate count, total coliforms, fecal coliforms and *E. coli*.

Pathogen isolation and identification is available for foods implicated in food borne illness outbreaks. Tests available include, but are not limited to, the following:

- *E. coli* O157:H7 detection and isolation
- *Salmonella* detection and isolation
- *Listeria* spp. detection and isolation
- *Campylobacter* spp. detection and isolation
- *Bacillus cereus* plate count
- Staphylococcal enterotoxins detection
Section 6: Foodborne Pathogens

Food samples are analyzed according to methods specified in the Bacteriological Analytical Manual (FDA BAM) Microbiology Laboratory Guide (USDA MLG) by methods specified by the Centers for Disease Control and Prevention (CDC), or the Food Emergency Response Network (FERN). When appropriate, rapid analytical test kits are used to screen samples for pathogens to provide quicker test results during food outbreak investigations or emergencies. The rapid test results usually take only 1 to 2 days. However, positive results of these tests are only presumptive and conventional tests need to be done to confirm these results.

Preliminary results are usually available within 48 to 72 hours after processing has begun. Confirmatory test results are usually available within 48 hours to ten days depending on the test organism. Please contact the Foodborne Pathogens Section at (602) 542-6130 at any time for updates on the progress of the testing. Generally, final reports are mailed out 3 to 11 days after initial processing begins.

Interpretation of lab results is the responsibility of the submitter. The laboratory will consult with the submitter, if requested. No legal food standards are available on most products, so care and common sense are needed in the interpretation of lab data. Use your experience and comparisons to evaluate the results.
Water Samples (performed by Analytical Preparation Services (APS))

The laboratory no longer accepts routine water samples for microbiological analysis. Samples are accepted from the County Health Departments with prior approval from management. The laboratory tests drinking water for the presence of coliforms and E. coli in compliance with the Safe Drinking Water Act. In addition, the laboratory tests surface or source waters, wastewater and runoff waters for indicator organisms. Please call the Arizona State Public Health Laboratory before submitting or shipping water samples for analysis.

Collection

Drinking Water Samples

Drinking water samples should be collected in sterile four-ounce whirl-pak bags or sterile collection bottles with sodium thiosulfate added to neutralize any chlorine in the water. Aseptically collect water from the sample tap. If using sterile collection bottles fill to the 100 mL line and leave adequate air space. If using the whirl-pak bags, collect 125 mL of water. Be sure to whirl them closed tightly and tie the tabs together securely.

Other Water Samples

Surface water, source waters, runoff waters, etc. can be aseptically collected in any appropriate size sterile whirl-pak bag or bottle (sodium thiosulfate is not needed); however, at least 125 mL is needed to test.

Shipment of Specimens

Drinking Water Samples

Drinking water samples must be received and tested within 30 hours of collection. It is recommended that samples arrive the first of the week whenever possible. Samples may be mailed or sent by courier to the Arizona State Public Health Laboratory to arrive the next day. While drinking water samples do not need to be iced during transit, it is recommended when feasible to cool samples. Each sample must be accompanied by a properly completed Water Microbiological Sample Submission Form. Information regarding how to obtain collection/submission supplies can be found in Section 9: Requesting Collection Kits and Mailing Containers. The form is also located on the website https://azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving-forms.

Other Water Samples

These waters need to be received in the laboratory within six hours of collection, and must be iced during transit. Since the transit time is so short, it is usually best to send the water samples to the laboratory by courier. A properly completed Water Microbiological Sample Submission Form must accompany each sample. More detailed information regarding how to obtain collection/submission supplies can be found in Section 9: Requesting Collection Kits.
Section 6: Foodborne Pathogens

and Mailing Containers. Before submitting these water samples, please call the Analytical Preparation Services Section at (602) 542-1147 to arrange for testing.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

**Drinking Water Samples**

Drinking water samples are routinely tested for the presence of total coliforms and *E. coli* using the enzyme substrate coliform test. This method provides results in 18 to 24 hours. This is the EPA approved method SM 9223B.

Results of drinking water coliform tests are usually available within 18 to 24 hours after processing has begun. All positive results are called to the submitter, providing that a telephone number has been supplied. In addition, all compliance positive results and repeat samples are faxed to ADEQ (Arizona Department of Environmental Quality). Leaked in transit and too long in transit samples are also called to the submitter. Final reports will usually be mailed one to two days after initial processing. If the sample is checked as a compliance sample, a copy is sent to the submitter and ADEQ.

Normally, the maximum contaminant level for total coliforms in drinking water is based on the presence or absence of coliform organisms in a 100 mL sample. A single water sample can have 0 coliforms per 100 mL. Other rules apply when more routine samples are collected, as the ADEQ compliance Department dictates. The number of samples required is based on the population served by a public water system. If a compliance sample is positive, repeat samples need to be collected. Please contact your ADEQ compliance officer to determine the number and location to collect these repeat samples.

<table>
<thead>
<tr>
<th>Method Name</th>
<th>Units</th>
<th>Standard Method Number</th>
<th>Holding Time</th>
<th>Matrix</th>
<th>Temp °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enzyme Substrate Coliform Test</td>
<td>Presence or Absence/100 mL</td>
<td>SM 9223B</td>
<td>30 Hours</td>
<td>Drinking, Well or Ground Water</td>
<td>Ambient</td>
</tr>
<tr>
<td>(Colilert/Colisure)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Colilert MPN - Most Probable Number (QuantiTray - MPN)</td>
<td>MPN Index per 100 mL</td>
<td>SM 9223B</td>
<td>8 Hours</td>
<td>Surface/Ambient and Wastewater</td>
<td>&lt; 10 °C</td>
</tr>
</tbody>
</table>

- Holding time of 30 hours for drinking water is the time of collection to start of incubation
- Holding time of 8 hours for surface/ambient and wastewater is time of collection to time of test start.
Section 7: Bioemergency Detection and Response
(Select Agents)

Since the terrorist events of September 2001, the ASPHL has set guidelines for the submission of miscellaneous powders and other suspicious substances for detection of priority biological agents (e.g., anthrax, plague, etc.). Additionally, clinical specimens of patients exposed to an intentional release of these priority biological agents, as well as patient specimens in association with naturally occurring infection, may be submitted for the organisms listed below. Notification of molecular results for the appropriate agent(s) will occur by telephone, usually within 2-5 hours of test completion followed later with final results based on organism recovery and biochemical assessment.

The Arizona Department of Health Services State Public Health Laboratory is the only Laboratory Response Network (LRN) reference laboratory in the State. As an LRN reference laboratory utilizing CDC developed procedures and testing algorithms, we have the ability to provide confirmatory rule-in/rule-out service for the priority biological agents: Bacillus anthracis, Brucella spp., Burkholderia spp, Coxiella burnetii, Francisella tularensis, Yersinia pestis, Ebola virus, MERS-CoV, and Orthopox viruses. Given that a number of the organisms listed have low (10-100 organisms) infectious doses we ask that extreme caution be taken when handling specimens suspected of containing any of these organisms. Brucella spp. and F. tularensis are consistently listed as some of the most commonly reported laboratory-associated bacterial infections.

Please contact the State Bureau of Epidemiology and Disease Control at (602) 364-3676 (main number) or (480) 303-1191 (after hour’s number) and the Bioemergency Detection and Response Laboratory at (602) 364-0999 before submitting samples for potential outbreak or unusual suspect organisms. In the event that an intentional release of any biological agent is suspected, contact your local county health department, local law enforcement agencies, and the FBI Phoenix field office at (602) 279-5511 to inform them of the incident.

Specimen Collection and Shipping

Collected samples, clinical or environmental, should be sent to the Arizona State Public Health Laboratory for testing as soon as possible to ensure the reliability of test results and/or to maximize the potential for recovery of viable organisms where appropriate. Refer to the American Society for Microbiology (ASM) Sentinel Laboratory procedures for acceptable specimen type and handling. Sample collection should be consistent with current medical practices for the disease/organisms biology. All clinical samples must have the following information on the submission form: Patient name, birth date, date of collection, sample source, contact information and test request.

Clinical samples, or reference isolates, should be submitted when a high suspicion exist that one of the organisms listed may be the cause of clinical disease (based on clinical signs, symptoms and exposure history) or when a recovered isolate from clinical or environmental samples cannot be ruled out based on traditional microbiological biochemical reactions. Additional information about appropriate clinical specimens and shipping conditions can be found on the ASM website listed below:
Environmental specimens should be of sufficient quantity and may consist of food, soil, smooth non-porous surfaces swab/wipe, powders, or packages (Table 1). Regardless of the sample type/source, collection should be performed by an individual with the knowledge and technical abilities to collect such a sample (i.e. food samples collected by trained sanitarian, powder samples collected by HAZMAT technician) and should be consistent with current practices for such sample. Prior to submitting environmental samples, contact the Bioemergency Detection and Response testing laboratory section at (602) 364-0999 for submission guidance. For details regarding the collection and submission of powder samples/suspicious unknowns including those associated with a threat, please contact your local law enforcement and refer to the Arizona Department of Health Service Suspicious Substances guidelines:

http://azdhs.gov/phs/emergency-preparedness/response-plans.htm

All organisms (in culture form) listed in this section are considered Category A infectious substances and must be shipped accordingly. For information regarding the packaging and shipping of Category A infectious agents please refer to “Section 8: Sample Submission Guidelines” of this manual and/or the ASM Packaging and Shipping Guidelines for Sentinel Laboratories

Table 1 – Environmental specimens

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Amount</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Smooth non-porous Surface (counter, instrument, etc.)</td>
<td>26 cm² (4 inch²) – Macrofoam swabs 645 cm² (100 in²) – Cellulose sponge-wipe 144 in² (929 cm²) - Gauze</td>
<td>Use sterile swab, Cellulose sponge-wipe or gauze. Synthetic fibers, synthetic or metal shafts strongly preferred.</td>
</tr>
<tr>
<td>Powder</td>
<td>Up to 5 g</td>
<td>Collect aseptically.</td>
</tr>
<tr>
<td>Food a</td>
<td>25 – 100 g</td>
<td>If food is not available, submit empty containers. Food must be suspected of being intentionally contaminated with one of the agents listed below.</td>
</tr>
<tr>
<td>Isolate a</td>
<td>Isolate plate or slant and original isolation plate</td>
<td>Send in both plates or tubes.</td>
</tr>
</tbody>
</table>

* Refrigerate immediately and transport on ice. Keep good records and send the Arizona State Public Health Laboratory a copy.
Results Reporting

The results of clinical and environmental samples will be reported to the submitter and all other relevant agencies in a manner (phone notification or electronic messaging) and time frame consistent with the submitted sample type and the desired organism identification algorithm. For questions regarding sample submission, turn-around-times or reporting mechanisms please contact the Bioemergency Detection and Response laboratory section at (602) 364-0999.

Following the detection and confirmation of a Select Agent (i.e. *B. anthracis*, *Brucella spp.*, *Burkholderia mallei/pseudomallei*, *C. Burnetii*, *F. tularensis*, *Y. pestis*) the Select Agent Program requires the submission of a APHIS/CDC Form 4A (Report of the Identification of a Select Agent or Toxin). Both the laboratory performing the confirmation/identification and the laboratory submitting the specimen for identification must complete designated sections of the Form 4. When a select agent is identified by ASPHL, the submitting laboratory will be contacted by ASPHL with the information regarding the need to fill out and submit the sections C and D of the APHIS/CDC Form 4A to CDC. A complete list of Select Agents can be found on the following website: [http://www.selectagents.gov/SelectAgentsandToxinsList.html](http://www.selectagents.gov/SelectAgentsandToxinsList.html)
Anthrax
*Bacillus anthracis*

Members of the *Bacillus* genus are aerobic, gram-positive spore forming bacteria. *Bacillus anthracis* is the causative agent of anthrax.

**Collection**

For cutaneous anthrax, sterile swabs are appropriate for collection of vesicular fluid and eschar material. Vesicular fluid should be obtained from a previously unopened vesicle(s) using a sterile swab. To collect eschar material, carefully lifting the eschar’s outer edge, insert a sterile swab and slowly rotate beneath the edge of the eschar without removing it. Place the swab in the appropriate transport container and ship at room temperature.

If intestinal or pulmonary anthrax is suspected, blood, serum, plasma, pleural fluid, transtracheal aspirates, sputum, fresh or frozen tissue, stool or rectal swab can be submitted for culture and/or PCR.

**Shipment of Specimens**

*Bacillus* species are hardy and usually survive transport to the ASPHL either in freshly collected specimens or in a standard transport medium. Specimens should be shipped/transported room temperature if they will not arrive at the ASPHL within 1 hour of collection. Reference isolates may be submitted on agar slants.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Cultures are checked daily for characteristic macroscopic morphology. Suspected isolates are tested biochemically and by Real-Time PCR. Confirmation is made by using LRN protocols and algorithms. Once a Bacillus isolate has been confirmed as negative for anthrax, any further identification will require approval from the Technical Supervisor.

All results, positive or negative will be phoned to the submitting agencies and when appropriate, reported to the Bureau of Epidemiology and Disease Control and local and federal law enforcement.
Brucellosis

*Brucella* spp.

*Brucella* spp. are small, non-motile gram-negative coccobacilli that are pathogenic to humans and animals. *Brucella* spp. are usually transmitted to humans by direct contact, consumption of contaminated food products, or inhalation. *Brucella* is one of the most common causes of laboratory acquired infections.

**Collection**

Specimens that can be collected and cultured for the isolation of *Brucella* include blood*, bone marrow, abscess fluid spleen and liver biopsies, cerebrospinal fluid (CSF) and joint fluid. Additionally, recovered isolates from clinical specimens and whole blood may be submitted for PCR analysis. Environmental samples such as food and water may be submitted.

*When brucellosis is suspected, multiple blood cultures should be obtained.*

**Shipment of Specimens**

Specimens should be cultured as soon as possible after collection or refrigerated if delays are unavoidable. Blood and body fluid samples should be shipped at 2-8 °C, on cold packs (wet ice if necessary). Tissues should be sent frozen at ≤-20 °C, e.g. on dry ice.

Reference isolates may be submitted on agar slants

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Cultures for *Brucella* are held for 14 days, and are checked daily for typical growth. Suspected isolates are examined by Gram staining for typical microscopic morphology. Identification, confirmation and speciation are made through biochemical testing and Real-Time PCR.

All results, positive or negative will be phoned to the submitting agencies and when appropriate, reported to the Bureau of Epidemiology and Disease Control and local and federal law enforcement.
Ebola

Under the use of an FDA Emergency Use Authorization (EUA) the ASPHL has the ability to perform PCR testing on whole blood, serum, plasma, and urine samples from patients suspected of being infected with Ebola virus. Because this test is offered under the FDA’s EUA, patients must meet specific clinical and/or epidemiological criteria and must have prior approval for Ebola testing from the Bureau of Epidemiology and Disease Control prior to sample submission. Current information on Ebola virus, including case definitions, is available at http://www.cdc.gov/vhf/ebola/index.html. Contact the Infectious Disease Section of the Bureau of Epidemiology and Disease Control at (602) 364-3676 (main number) / (480) 303-1191 (after hour’s number) for additional information and request for testing services.


Acceptable samples include:

- Whole blood
- Serum
- Plasma
- Urine

NOTE: Urine should not be the sole specimen tested from a patient. If a urine specimen from a patient is tested, it should be tested alongside a blood specimen from the patient.

Specimen Handling and Storage

- Whole blood can be stored for up to 7 days at 2 – 8 °C prior to extraction.
- Serum, plasma and urine may be frozen if a delay in extraction is anticipated. Specimens should be frozen at ≤ -70 °C, if available.
Middle East Respiratory Virus, MERS-CoV

Under the use of an FDA Emergency Use Authorization (EUA) the ASPHL has the ability to perform PCR testing on respiratory specimens from patients suspected of having Middle East Respiratory Syndrome. Because this test is offered under the FDA's EUA, patients must meet specific clinical and/or epidemiological criteria and must have prior approval for MERS-CoV testing from the Bureau of Epidemiology and Disease Control prior to sample submission. Refer to Interim Guidelines for Collection, Handling and Testing of Clinical Specimens from Patients Under Investigation (PUIs) for Middle East Respiratory Syndrome Coronavirus (MERS-CoV) (http://www.cdc.gov/coronavirus/mers/guidelines-clinical-specimens.html) for information on specimen collection and handling. Contact the Infectious Disease Section of the Bureau of Epidemiology and Disease Control at (602) 364-3676 (main number) / (480) 303-1191 (after hour’s number) for additional information and request for testing services.

Acceptable samples include:
- Nasopharyngeal
- Oropharyngeal
- Sputum
- Lower respiratory tract aspirates/washes
- Serum

Specimen Handling and Storage

Specimens can be stored at 2 – 8 °C for up to 72 hours after collection. If a delay in sample processing is expected, store the specimen at -70 °C or lower.
Melioidosis and Glanders

*Melioterla* spp.

Members of the genus are slightly curved gram-negative bacilli. *B. mallei* is the causative agent of glanders and *B. pseudomallei* the causative agent of melioidosis. Laboratory-acquired infections have been documented, thus all patient specimens and culture isolates suspected of containing/being either *B. pseudomallei* or *B. mallei* should be handled while wearing gloves and a gown. If either of these organisms is suspected further work should be done in a BSL-3 setting or at minimum BSL-2 facilities with BSL-3 practices.

Laboratories should not accept environmental or animal specimens; such specimens should be forwarded directly to the ASPHL.

**Collection**

Specimens that can be collected and cultured for the isolation of *B. mallei* and *pseudomallei* include blood, urine, sputum, abscesses and tissue aspirates. For PCR analysis, submit whole blood, serum, or isolates (pure cultures or primary growth plates).

**Shipment of specimens**

Specimens should be cultured as soon as possible after collection or refrigerated if delays are unavoidable. Blood and bone marrow samples should be collected directly into an appropriate blood culture bottle and transport at room temperature as soon as possible. For respiratory (sputum or bronchoscopically obtained specimens) and urine specimens transported with 2-24 h after collection, these samples should be store and transport at 2-8 °C.

See Section 8: Sample Submission Guidelines

**Reporting and Interpretation of Results**

Cultures for *Burkholderia* spp. are held for 10 days and checked daily for typical growth. Suspected isolates are examined by Gram staining for typical microscopic morphology. Identification is made by Real-Time PCR and biochemical testing. Positive isolates of *B. mallei* or *B. pseudomallei* may be forwarded to the Centers for Disease Control and Prevention in Atlanta, Georgia for confirmation of laboratory results.

All results, positive or negative will be phoned to the submitting agencies and when appropriate, reported to the Bureau of Epidemiology and Disease Control and local and federal law enforcement.
Orthopoxvirus

Orthopoxviruses are one of 8 genera that comprise the Poxviridae family of viruses and includes viruses such as variola virus (Smallpox), vaccinia virus (Smallpox vaccine), and monkeypox. The ASPHL conducts testing to detect the presence of orthopoxviruses as a means to rule-out the possibility of Smallpox.

Collection

Prior to sample collection and shipment, if Smallpox is suspected a mandatory CDC Risk Assessment algorithm must be completed by Arizona State Department of Health Services personnel. In order for the department to evaluate each individual case per the CDC algorithm, contact the Bioemergency section at (602) 364-0999 (main line) / (480) 303-1676 (after hours) at the ASPHL or the Infectious Disease Section of the Bureau of Epidemiology and Disease Control at (602) 364-3676 (main number) / (480) 303-1191 (after hours number) with a list of relevant clinical symptoms and complete patient information including vaccination and travel histories.

If testing is to be conducted at the ASPHL, the following samples may be submitted for testing: vesicular fluid, skin or crust form the roof of a vesicle, nylon swab of a lesion, or fresh tissue biopsy (Submit swabs, biopsy tissue and scabs dry, DO NOT add viral transport medium, a dry swab is preferred.).

Caution should be used when collecting clinical specimens thought to contain Smallpox. All processes including collection, processing, and packaging and shipping should be performed using BSL-2 (or BSL-3 if available) practices. The individual collecting the sample should wear the appropriate personal protective equipment including gloves, disposable gown, shoe covers, mask and eyewear or face shield. Respiratory protection is not necessary, but is recommended for individual with recent vaccination.

Contact the Bioemergency Detection and Response Laboratory (602) 364-0999, for details or questions regarding the specimen collection process.

Shipment of Specimens

If upon completion of the risk assessment it is decided that the sample meets the CDC criteria for Smallpox testing, the Arizona State Department of Health Services State Public Health Laboratory will either accept and test the sample or forward the specimen to a laboratory with the appropriate safety level facilities for testing.

Package specimens from each individual being tested separately, do not package samples from multiple patients in one bag. Samples should be shipped within 24 hours of collection and be held at 2 – 8 °C. If samples will not be received in the lab within 24 hours, samples should be stored and shipped on dry ice or at -20 °C to -70 °C. All packages must meet the current IATA and DOT standards for shipping infectious substances.

See Section 8: Sample Submission Guidelines.
Reporting and Interpretation of Results

Specimens submitted for Smallpox testing will be tested for the presence of orthopoxvirus DNA by PCR. Testing conducted at the ASPHL detects the presence of orthopoxvirus DNA but does not specifically detect the presence of Smallpox DNA.

All results will be reported (via phone call) to the submitting agencies and the Bureau of Epidemiology and Disease Control. Positive results will be reported to Centers for Disease Control and Prevention. Positive sample material may be forwarded to the Centers for Disease Control and Prevention in Atlanta, Georgia for additional laboratory testing.
Plague

*Yersinia pestis*

*Yersinia* spp. are non-spore-forming, gram-negative, coccobacilli or rod shaped bacilli. *Yersinia pestis* is the causative agent of the Plague. Plague can present in one of three clinical manifestations: bubonic, septicemic or pneumonic. Initially humans are infected with the bite of an infected animal or a flea. Once bitten, the affected individual could develop bubonic or septicemic plague from one of these bites. This disease development could occur with or without the development of pneumonic plague. If the disease progresses to the lungs, patients can transmit *Y. pestis* via aerosols to others. Patients with plague often develop necrotic lesions in the peripheral blood vessels, which can give the skin a black color, thus the name “black death”.

**Collection**

Clinical samples that may be submitted to the laboratory for identification of *Yersinia pestis* include aspirates (i.e. lymph node), biopsy of affected area (e.g. lymph node, lung), bronchial wash, transtracheal aspirate, blood, and sputum. Autopsy specimens include: abscess material or sections of lymph node, lung, liver, spleen.

Animal specimens may also be sent for isolation and identification procedures. Lymph node abscess material and necropsy specimens including: (abscess material or sections of lymph node, lung, liver, spleen or bone marrow scrapings may be used for animal submissions.

**Shipment of Specimens**

Transport samples to the ASPHL as soon as possible. Store samples containing suspected *Y. pestis* at 2-8°C to maintain viability and process them as quickly as possible. If processing is delayed, store specimens (except tissue samples) for culture in glycerol containing solutions (10% final concentration) at ≤-70 °C and ship on dry ice. Tissue samples can be directly frozen at ≤-70 °C and shipped on dry ice.

Tissue samples such as buboes, lung or lymph nodes should be collected into a sterile container. For small samples, add 1-2 drops of sterile normal saline to keep the tissue moist.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

Cultures are identified by observing typical colonial morphology. Typical colonies are presumptively identified using a Direct Fluorescent Antibody (DFA) test and/or Real-Time PCR. A positive DFA or PCR test is considered presumptive positive for *Y. pestis*. All presumptive positive results are telephoned to the submitting agency and to the Vector Borne and Zoonotic Disease Section of the Bureau of Epidemiology and Disease Control. Cultures suspected of containing Plague are tested and confirmed using conventional biochemicals.

Cultures are held for 7-10 days before reporting as negative.
Q Fever
Coxiella burnetii

Q fever is a zoonotic disease caused by Coxiella burnetii, a species of bacteria that is distributed globally. Cattle, sheep, and goats are the primary reservoirs of C. burnetii. C. burnetii is an intracellular bacterium that must be grown in cell culture.

Collection

Specimens that can be collected and tested by the ASPHL are whole blood in EDTA and environmental swabs.

Shipment of specimens

Specimens should be collected as soon as possible and refrigerated if delays are unavoidable.

All specimens should be kept at refrigerated temperatures during shipment.

See Section 8: Sample Submission Guidelines.

Reporting and Interpretation of Results

Whole blood and environmental swabs are tested by Real-time PCR for presence of C. burnetii DNA. Positive samples of C. burnetii will be reported and may be forwarded to the Centers for Disease Control and Prevention in Atlanta, Georgia for confirmation of laboratory results.
Tularemia

*Francisella tularensis*

The genus *Francisella* are tiny aerobic non-motile gram-negative coccobacilli. *Francisella tularensis* is the causative agent of tularemia, a disease that can be misdiagnosed early in the infection because the symptoms are not unique. Humans typically acquire *F. tularensis* after contact with tissues or body fluids of infected animals or an insect bite.

**Collection**

During infection, direct isolation is achieved from ulcer scrapings/swabs, lymph node biopsies, bronchial/tracheal washings, sputum, and pleural fluid. In human cases, several sources should be considered. Organisms are invariably present in significant numbers in fluid from obvious local lesions. Skin around the lesion should be cleansed with alcohol and allowed to dry before opening the papule and exposing the fluid. Organisms may persist for long periods of time in lymph nodes and may be isolated by node biopsy.

**Shipment of Specimens**

Transport samples to the ASPHL as soon as possible. Store samples without preservatives (formaldehyde, alcohol), at 2-8°C (not frozen) prior to processing. Freezing of samples, unless in a preservative environment, such as tissue specimens or glycerol containing solutions (10% final concentration), is not recommended because of lysis of live bacteria upon thawing. Transport blood samples directly to laboratory at room temperature. Hold at room temperature, do not refrigerate. Submit tissue, scraping, or aspirate in a sterile container, for small tissue samples, add several drops of sterile normal saline to keep the tissue moist. Transport specimen chilled (2-8°C).

Swabs: Obtain a firm sample of the advancing margin of the lesion. If using a swab transport carrier, the swab should be reinserted into the transport package and the swab fabric moistened with the transport medium inside the packet. Transport at 2-8°C.

See Section 8: Sample Submission Guidelines.

**Reporting and Interpretation of Results**

*F. tularensis* requires cysteine enriched medium for growth. The historic medium of choice is cysteine glucose blood agar; however any media containing cysteine supplementation is acceptable. Cultures are observed for 7-10 days before reporting as negative. Cultures are observed for typical colonial morphology. Suspect colonies are checked microscopically by Gram staining, where they appear as faintly staining gram-negative coccobacilli. Confirmation of the isolate is determined by Direct Fluorescent Antibody, and Real-Time PCR.

All positive cultures are reported to the submitting agency and the Vector-Borne and Zoonotic Disease Section of the Bureau of Epidemiology and Disease Control. *F. tularensis* isolates are forwarded to the Centers for Disease Control and Prevention at Atlanta, GA. upon request.
Section 8: Sample Submission Guidelines

Submit all samples to the following location:

Arizona Department of Health Services
State Public Health Laboratory
250 N. 17th Ave
Phoenix, AZ 85007
(602) 542-1188

All infectious material must be classified as either Category A (UN2814) or B (UN3373) and must be transported to the Arizona State Public Health Laboratory according to appropriate IATA (International Air Transportation Association), USPS (United States Postal Service) and DOT (U.S. Department of Transportation) regulations. The list of Category A organisms as outlined by IATA is contained at the end of this section. All infectious material must be triple-packaged to protect against breakage and/or leakage during transportation. An ASPHL Submission Form must accompany every sample submitted for testing, the form is available at:


Category B shipments must follow Packaging Instruction (PI) 650. Category A shipments must follow PI 620 and shipments with dry ice must follow PI 954. All samples and their containers must be identified with the appropriate labels, client and patient information.

Any samples which are leaking and/or not properly identified will be rejected. The following are brief guidelines for properly triple-packaging and shipping specimens for infectious testing at the Arizona State Public Health Laboratory:

- **Primary Container**
  - Must be securely sealed; leak-proof for liquids and silt-proof for solids
    - NOTE: screw caps and parafilm recommended
    - NOTE: Primary OR secondary container must be pressure and temperature capable (95kPa) if air transportation is used
  - Samples must be properly labeled with patient identifying information
    - Specimen primary containers for Mycobacterial examination must be labeled with the patients name, specimen type, date AND time of the collection.
  - For Category A the maximum quantity for a cargo plane is 4 L or 4 kg. For a passenger plane the maximum quantity is 50 mL or 50 g
  - For Category B, the maximum quantity is 500 mL or 500 g
  - Wrap with absorbent material sufficient for entire contents, and cushioning material

- **Secondary Container**
  - Securely sealed and watertight/leak-proof
    - NOTE: Primary OR secondary container must be pressure and temperature capable (95kPa) if air transportation is used
    - NOTE: If you have the appropriate materials you can place multiple primary containers inside a secondary container
A completed itemized list of contents must be placed outside of, or surrounding the secondary container
  - NOTE: An ASPHL Submission Form will satisfy the list of contents requirement
  - Place absorbent and cushioning material between the primary and secondary containers
  - Affix a biohazard symbol to the secondary container

- **Tertiary/Outer Container for CATEGORY B shipments**
  - Outer package must be rigid and of good quality
  - Affix UN3373 Biological Substance, Category B diamond shaped hazard label.
    - Do NOT affix biohazard symbol to outer package
  - Full name, complete address and phone number of shipper (responsible person).
  - Full name, complete address and phone number of recipient

- **Tertiary/Outer Container for CATEGORY A shipments**
  - Outer package and inner containers must be UN certified, outer package must contain the UN symbol
  - For Category A shipments containing infectious material affecting humans, affix **UN2814 Infectious Substances, Affecting Humans** diamond shaped hazard label
  - For Category A shipments containing infectious material affecting animals, affix **UN2900 Infectious Substances, Affecting Animals** diamond shaped hazard label
    - NOTE: A list of UN2814 and UN2900 organisms is contained at the end of this section
    - NOTE: if the infectious material affects both humans and animals, then treat as UN2814
  - Orientation marks (up arrows) must be present on two (2) sides of outer box
  - Full name, complete address and 24 hour direct phone number of shipper (responsible person)
  - Full name, complete address and phone number of recipient
  - NOTE: Make sure to write 6.2 above the UN2814 of the label. Leave the technical name off of the box.
  - NOTE: You must include the full technical name of the suspected unknown or Select Agent on the Dangerous Goods Form placed in with the Secondary Container
  - For Infectious Substance and Dry Ice Label template examples, refer to the ASPHL Shipping/Receiving website

- **Additional Documentation and Considerations**
  - **Temperature Considerations**
    - Consult appropriate sections within this *Guide to Laboratory Services* document for specific shipping temperatures based on the organism or laboratory section performing test
    - If wet ice or ice packs are to be used for maintaining refrigerated shipping temperatures ensure there is sufficient absorbent material contained within to absorb all moisture if ice melts during transit so integrity of box is not compromised.
NOTE: It is recommended to place wet ice and/or ice packs inside a zip-lock bag and surround this with absorbent material. If dry ice is to be used to maintain sub-frozen temperatures ensure that the package conforms to PI 954 and that dry ice is not placed inside any tightly sealed container that will prevent the release of carbon dioxide gas during sublimation.

NOTE: Dry ice will degrade rapidly therefore it must be purchased, obtained and used as close to actual shipping as possible.

**Dangerous Goods Shipper's Declaration**
- A Shipper’s Declaration must accompany all Category A shipments
  - NOTE: A minimum of 3 color and signed copies is needed
- A Shipper’s Declaration is not needed for Category B shipments
- A Shipper’s Declaration is not needed if only shipping dry ice, or dry ice with a Category B shipment

**Select Agent and Toxin Transfers**
- Shipping of any known Select Agent or Toxin must have prior approval and a completed CDC/APHIS Form 2.
- Any “suspected” Select Agents must be shipped as either Category A or Category B as designated by its classification.
- For additional information please visit the Select Agents website at: [http://www.selectagents.gov/form2.html](http://www.selectagents.gov/form2.html) or contact the Arizona State Public Health Laboratory Bioemergency Response Section at (602) 364-0999 for further assistance.

**Training**
- Anyone who packages or ships infectious material must receive appropriate training. There are several “hands-on” and online courses and trainings available. For further information or the next scheduled course please contact the ASPHL Technical Trainer at (602) 542-6175.

**Supplies**
- The Arizona State Public Health Laboratory offers several collection kits and materials for submitting samples. Please see Section 9 for further information.

**Regulations and Additional Guidance**
- Arizona State Reporting and Isolate Submission Requirements A.A.C. R9-6-204  [http://azdhs.gov/labreporting](http://azdhs.gov/labreporting)
- United States Postal Service (USPS) 39 CFR Part 20, International Postal Service (International Mail Manual), and Part 111, General Information on Postal Service (Domestic Mail Manual). Regulations on transporting...
infectious substances through the USPS are codified in Section 601.10.17 of the Domestic Mail Manual and Section 135 of the International Mail Manual. A copy of the Domestic and International Mail Manuals may be obtained from https://about.usps.com/manuals/welcome.htm or the Government Printing Office by calling Monday through Friday, 7:30 a.m. - 9:00 p.m. EST: 202-512-1800, 866-512-1800 (toll free), or from:
http://www.gpo.gov

  http://www.osha.gov/

- Technical Instructions for the Safe Transport of Dangerous Goods by Air (Technical Instructions). International Civil Aviation Organization (ICAO). A copy of these regulations may be obtained from the ICAO Document Sales Unit at (514) 954-8022, Fax: (514) 954-6769, E-Mail: sales_unit@icao.int, or from: http://www.icao.int

- Dangerous Goods Regulations International Air Transport Association (IATA). These regulations are issued by an airline association, are based on the ICAO Technical Instructions, and are followed by most airline carriers. A copy of these regulations can be obtained from: http://www.iata.org/Pages/default.aspx or http://www.who.int/en/

- Please contact the Arizona State Public Health Laboratory (ASPHL) for appropriate specimen types and shipping instructions for specimen referral to the Centers for Disease Control and Prevention (CDC) for non-routine testing not offered at the ASPHL.
The following list is not exhaustive. It is the list of Category A organisms as outlined by the IATA regulations. If there is any doubt as to whether the shipment should be sent as Category A or B, please contact the Arizona State Public Health Laboratory at (602) 542-1190 or (602) 364-0999 for assistance.

### IATA Category A Organisms

<table>
<thead>
<tr>
<th>UN 2814</th>
<th>Infectious substances, affecting humans</th>
</tr>
</thead>
<tbody>
<tr>
<td>Micro-Organism</td>
<td>UN 2814</td>
</tr>
<tr>
<td><strong>Bacillus anthracis</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Brucella abortus</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Brucella melitensis</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Brucella suis</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Burkholderia mallei-Pseudomonas mallei-Glanders</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Burkholderia pseudomallei-Pseudomonas pseudomallei</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Chlamydia psittaci</strong> (avian) (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Clostridium botulinum</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Coxiella burnetii</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Crimean-congo hemorrhagic fever virus</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Dengue virus</strong> (cultures)</td>
<td></td>
</tr>
<tr>
<td><strong>Eastern equine encephalitis virus</strong> (cultures only)</td>
<td></td>
</tr>
<tr>
<td><strong>Escherichia coli</strong>, verotoxigenic (cultures only)</td>
<td></td>
</tr>
</tbody>
</table>
Section 8: Sample Submission Guidelines

Category A Shipping Examples

Illustrations of proper shipping procedures for Category A samples, including primary and secondary packaging, labeling requirements, and cross-section diagrams of properly packed samples.
Category B Shipping Examples

A completely labeled outer package. The primary container inside the package contains a Biological Substance, Category B substance and is packed according to PI 650.
Category B Checklist

Sample ID: ____________________________
Packager Name/Initial: ____________________
Date: __________________

**CATEGORY B CHECKLIST**
- UN 3373 Biological Substances, Category B
- IATA Packing Instruction (PI) 650
- FedEx, UPS, USPS (US Mail), private couriers

**Primary:**
- Specimen properly labeled with patient ID information
- ≤50mL or ≤50g maximum quantity
- Securely sealed & watertight/leakproof (screw cap receptacle and paraffilm)
  Note: a Petri dish is not an acceptable primary container
- Wrapped in absorbent material sufficient for entire contents
- Wrapped in cushioning material (bubble wrap)
  - Primary OR secondary container pressure and temperature capable (95kPa)

**Secondary:**
- Securely sealed and watertight/leakproof
- Primary OR secondary container pressure and temperature capable (95kPa)
- A completed itemized list of contents (requisition or sample submission form) is placed between the secondary packaging and the outer packaging (NOT inside the secondary packaging)
- Absorbent material is placed between the primary and secondary packaging
- Biohazard symbol on secondary package required if shipping via US Mail (USPS)
- (Optional) Additional cushioning material placed between primary and secondary

**Outer Package (Rigid):**
- Package is rigid and of good quality (acceptable to reuse Category B packages)
- UN 3373 Biological Substances, Category B diamond shaped label
- Quantity of infectious material is listed
- Quantity of sample – volume (mL) or weight (g)
- Must not contain more than 4 L, 4000mL or 4kg
- Do NOT put biohazard symbol on outer package
- Full name, complete address and phone number of person responsible for the shipment
  (This can be either the shipper or the recipient, but must be someone knowledgeable of the contents)
- Full name, complete address and telephone number of the shipper
- Full name, complete address and telephone number of the consignee/recipient

**NOTE:** A Shipper’s Declaration is not needed for Category B samples OR if dry ice is used.

If dry ice is used consult dry ice shipping checklist
If overpack is used consult overpack shipping checklist
Dry Ice Checklist

Sample ID: ____________________________
Packager Name/Initial: ____________________________
Date: ________________

DRY ICE CHECKLIST
Combined with Category A, B, or Exempt Shipments
UN 1845 -Miscellaneous Hazard Class 9
Packing Instruction (PI) 954 “formerly PI 904”

☐ Properly ventilated package
   NOTE: Does not have to be a UN certified package
☐ Overpack sticker/label needed for Category A shipments only
   (Category B packages will be packaged differently than A)
☐ The net quantity of dry ice used in kg is listed
☐ The quantity of the dry ice per package is less than 200kg
☐ Irrelevant masks and labels removed from package
☐ The UN number “UN 1845” label or sticker
☐ Miscellaneous Hazard Class 9 label or sticker
☐ Full name, complete address and phone number of person responsible for the shipment
   (This can be either the shipper or the recipient, but must be someone knowledgeable of the contents)
☐ Full name, complete address and telephone number of the shipper
☐ Full name, complete address and telephone number of the consignee/recipient

Shipper’s Declaration for Dry Ice
Shipper’s Declaration must be completed for shipments containing Category A substances only, dry ice
alone does not require a Shipper’s Declaration

☐ The words “Carbon dioxide, solid” or “Dry ice” is contained on Shipper’s Declaration
   Recommended to use “Dry Ice”
☐ Packing group III
☐ Packing Instruction 954

Consult Checklist for Shipper’s Declaration for complete list
Section 9: Requesting Collection Kits and Mailing Containers

Supplies ordered from the Arizona State Public Health Laboratory (ASPHL) are to be used ONLY to submit specimens to the ASPHL. There are two Requests for Materials forms currently in use: a Newborn Screening Supplies Request Form and a Request Form for all other supplies available from the ASPHL. Supplies can be requested by faxing, emailing or calling the Receiving Section. All request forms are available as fillable or printable documents at:

https://www.azdhs.gov/preparedness/state-laboratory/index.php#shipping-receiving-forms

Arizona Department of Health Services  
Bureau of State Laboratory Services  
ATTN: Receiving Section  
250 North 17th Avenue  
Phoenix, AZ 85007

Fax: (602) 364-0758  
Phone: (602) 542-1190  
Email: labreceiving@azdhs.gov

Please request materials before they are required as the expected turnaround time per order is FIVE business days. Most materials do have a limited shelf life; therefore, only order what will be used before the expiration date. Please do not use expired kits or any kits in which the medium has changed characteristics. Dispose of the media properly and order replacement supplies. The following table provides information regarding submission forms, kit contents and expiration period of each kit. Submitters may use the Request for Materials form to order entire kits, as well as individual components.  
https://www.azdhs.gov/preparedness/state-laboratory/#shipping-receiving-forms

<table>
<thead>
<tr>
<th>KIT</th>
<th>CONTENTS</th>
<th>SHELF LIFE</th>
</tr>
</thead>
</table>
| Enteric Kit    | Instruction Sheet  
                 | Baggie  
                 | Media: Cary Blair  
                 | Store +20 to +25 °C | 1-2 years |
| Influenza Kit  | Instruction Sheet  
                 | N/P Swab  
                 | Media: Universal Transport Medium  
                 | Store +2 to +25 °C | 1-2 years |
| Pertussis Kit  | Instruction Sheet  
                 | N/P Swab  
                 | Media: Regan Lowe.  
                 | Store +2 to +8 °C | 4-6 months |
| Tuberculosis Kit | Sputum Vial  
                         | Metal Container  
                         | Cardboard Mailer  
                         | Store +20 to +25°C |
Section 9: Requesting Collection Kits and Mailing Containers

All Submission Forms can be obtained at:

https://www.azdhs.gov/preparedness/state-laboratory/#shipping-receiving-forms

The ASPHL requests that facilities submitting samples/specimens to the laboratory complete the submission forms electronically. The electronic completion of submission forms benefits everyone through ensuring that all information needed is completed, reducing interruptions to submitters, and improves the overall efficiency of the submission of samples/specimens.