<table>
<tr><th colspan="2">Louisiana Office of Public Health Laboratories</th></tr>
<tr><td>Test Name</td><td>Shiga Toxin Positive Escherichia coli (STEC)</td></tr>
<tr><td>PHL Location</td><td>Office of Public Health Laboratory Baton Rouge</td></tr>
<tr><td>CPT Code</td><td>87798 X 4</td></tr>
<tr><td>Synonyms</td><td>EHEC, STEC, Ecoli Toxin, O157</td></tr>
<tr><td>Brief Description of Test</td><td>This test is available when submitters get shiga toxin positive results from stool or GN/MAC broth and they are unable to isolate the toxin producing strain of E. coli.

Isolation and identification of Shiga toxin producing E. coli from Toxin positive stool or mixed culture broth.

Conventional PCR for :
E. coli mal-B promoter region
E. coli nt 393-651 of rfbE (0157:H7)
E. coli nt 454-633 of A subunit coding region of stx1
E. coli nt 603-857 of A subunit coding region of stx2</td></tr>
<tr><td>Possible Results</td><td>To determine E. coli O157 from non-O157 and to determine if E. coli of any kind is detected
  E. coli Detected or Not Detected
  O157 Detected or Not Detected

To determine if genes for shiga-toxin production are present
  Stx1 Detected or Not Detected
  Stx2 Detected or Not Detected

Inconclusive – No Culture Available for Confirmation</td></tr>
<tr><td>Reference Range</td><td>Not Detected</td></tr>
<tr><td>Specimen Type</td><td>
<table>
<tr><th>Specimen Container*</th><th>Specimen Type</th><th>Requested Volume (mL)**</th><th>Storage / Transport Temperature (°C)***</th><th>Acceptable Age or Condition of Specimen</th><th>Special Instructions</th></tr>
<tr><td>Media Tube for Slant</td><td>Stool Isolate</td><td>N/A</td><td>2-37°C</td><td>N/A</td><td>Specimens for 0157 testing would be routed to General Bacteriology. Specimens for STEC testing are routed to Molecular Biology.</td></tr>
<tr><td rowspan="2">Media Tube for Liquid</td><td rowspan="2">Stool - GN or MAC broth</td><td rowspan="2">1 mL</td><td>2-37°C</td><td>23 days</td><td rowspan="2">GN/MAC broth or isolate shipping of an STEC positive specimen requires Cat A shipping.</td></tr>
<tr><td>2-8°C</td><td>12 days</td></tr>
<tr><td>Cary Blair Transport</td><td>Stool in Transport</td><td>N/A</td><td>2-8°C</td><td>6 days</td><td>Cary Blair is only accepted with a patient matched isolate or broth.</td></tr>
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Note: Cary Blair liquid transport such as Remel R21610, not transport swabs.</td></tr>
<tr><td>Specimen Container(s):</td><td>Leak proof, screw cap slants/tubes.
Do not send inoculated agar plates through the mail system.</td></tr>
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Minimum volume accepted:	See specimen Type above.
Collection Instructions	Label specimen with Patient Name and a 2nd Unique Identifier such as a chart number or medical record number. DOB is not considered unique. Complete a LAB Form 93 to accompany the sample. Lab submission form must be thoroughly completed with patient's first and last name, 2nd patient identifier, gender, date of birth, date and time of collection, specimen source, test requested, submitter's name, address, fax and contact number. Additional information regarding patients' address is requested. Transport specimen to laboratory as soon as possible after collection/incubation. Keep submission forms insulated from specimens.
Storage and Transport Instructions	Stool – Follow Cary Blair manufacturer instructions for sample collection. Can be stored and shipped at 2-8°C for delivery within 6 days. Must be submitted with a patient matched broth or isolate. Gram Negative/MacConkey Broth – Incubate prior to shipment. Can be stored and shipped at 2-8°C for delivery within 12 days or 2-37°C for delivery within 23 days. Isolate in media slant – Incubate prior to shipment. Can be stored and shipped at 2-37°C. Specimens received outside of acceptable criteria will be resulted and reported to the submitter as Unsatisfactory. The state laboratory may still attempt STEC isolation from Unsatisfactory samples for surveillance reporting to Infectious Disease Epidemiology and/or CDC. Send sample to the Office of Public Health Laboratory Baton Rouge, 1209 Leesville Avenue, Baton Rouge, LA 70802 United Nations regulations (Division 6.2, Infectious Substances) stipulate that a verotoxigenic *E. coli* culture is a **category A (United Nations number 2814) infectious substance**, which is an infectious substance in a form capable of causing permanent disability or life-threatening or fatal disease in otherwise healthy humans or animals when exposure to the substance occurs. The International Air Transportation Association (IATA) and Department of Transportation (DOT) have modified their shipping guidance to comply with this requirement. Therefore, all possible and confirmed O157 STEC and non-O157 STEC isolates and Shiga toxin--positive broths should be shipped as category A infectious substances. If the identity of the infectious material being transported has not been confirmed or is unknown, but the material might meet the criteria for inclusion in category A (e.g., a broth culture that is positive for Shiga toxin or a stool culture from a patient that might be part of an O157 STEC outbreak), certain IATA regulations apply. Both IATA and DOT require that all persons who package, ship, or transport category A infectious substances have formal, documented training every 2 years.

Causes for Rejection	• Received outside acceptable transport/storage conditions • Improper labeling • Incorrect source • Cary Blair submitted without patient matched broth or isolate
Limitations of the Procedure	PCR results should not be used as a sole test for sample identification. Negative test results may occur from improper specimen collection, handling or storage, presence of inhibitor or because the number of organisms in the specimen is below the analytical sensitivity of the test. Unless the specimen is culture, a positive test result does not necessarily indicate the presence of viable organism. Escherichia coli and Shigella spp. are difficult to differentiate with molecular methods and both may produce shiga toxin. Conventional biochemical culture confirmation is suggested. The Molecular laboratory has the capability to perform PCR for shiga toxin 1 and shiga toxin 2. We use this PCR to help identify non-O157 STEC and to isolate the non-O157 STEC from other *E. coli* normally found in stool samples. Interpretation of Final Results—Several tests for microbiology laboratories are available for the detection of STEC, and they may be used alone or in combination. No testing method is 100% sensitive or specific, and the predictive value of a positive test is affected by the patient population that a particular laboratory serves. Specificity and sensitivity might be increased by using a combination of tests. However, when test results conflict, interpretation might be difficult, especially when your result and our result are compared. Discordant results (e.g., positive immunoassay at your laboratory but negative PCR result at our laboratory) might need to be discussed among the treating physician, public health epidemiologist and both hospital and public health laboratory staff; however, the outcome of most patient illnesses (i.e., resolution of symptoms or progression to HUS) is already known by the time discordant laboratory findings are resolved.
Interfering Substances	Toxin produced by *Shigella dysenteriae* type I
References	Candrian, U., Furrer, B., Hofelein, C. Meyer, R., Jermini, M., and Luthy, J. (1991) Detection of Escherichia coli and Identification of Enterotoxigenic Strains by Primer Directed Enzymatic Amplification of Specific Sequences. International Journal of Food Microbiology 12, 339-352 Paton, A.W., and Paton, J.C. (1997) Detection and Characterization of Shiga Toxigenic Escherichia coli by Using Multiplex PCR Assays for stx1, stx2, eaeA, Enterohemorrhagic E. coli hlyA, rfb-O111, and rfb-O157. Journal of Clinical Microbiology 36 (2), 598-602 R.F Wang et al. 1997. A Universal Protocol for PCR detection of 13 species of foodborne pathogens in foods. Journal of Applied Microbiology 83:727-736

	MMWR – Recommendations for Diagnosis of Shiga Toxin Producing *Escherichia coli* Infections by Clinical Laboratories (October 16, 2009)
Additional Information	Primers for E. coli tests were developed by AW Paton and JC Paton 1998 and Candrian et al 1991. Assay performance characteristics have been determined by the Louisiana Office of Public Health Central Lab. This test has not been cleared or approved by the U.S. Food and Drug Administration.
Release Date	03/28/2018
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