The CDC’s National Enteric Reference and Outbreak Laboratory Quality Assurance Panel (NERO QA Panel) for the identification and subtyping of bacterial enteric pathogens

2018 Summary Report

The CDC’s National Enteric Reference and Outbreak Laboratory Quality Assurance Panel (NERO QA Panel) for the identification and subtyping of bacterial enteric pathogens is organized and conducted by the Enteric Diseases Laboratory Branch, Centers for Disease Control and Prevention (CDC). The program included four panels that assess participants’ abilities in the following areas:

*Campylobacter* (5 isolates)
  - Phenotypic identification
  - Molecular identification

*Escherichia/Shigella* (5 isolates)
  - *Escherichia*:
    - Identification to the species level
    - For Shiga toxin producing *E. coli*:
      - O Antigen determination (top seven)
      - H Antigen determination (optional)
      - Virulence profiling
  - *Shigella*:
    - Identification to the subspecies level
    - Serotyping (as complete as possible)
    - Virulence profiling

*Salmonella* (5 isolates)
  - Identification to the subspecies level
  - Serotyping (phenotypic and/or genetic methods)

Other *Enterobacteriaceae* (2 isolates)
  - Identification to the species level

Confidential reports for each laboratory were issued via email between May and August 2018; this is a summary of the group’s performance on each of the panels. We are pleased with the participation we received in the 2018 cycle and gratefully acknowledge the support of the Public Health Agency of Canada, National Microbiology Laboratory, Winnipeg, Canada, who served as the referee laboratory.
**2018 Campylobacter QA Program Summary**

Fifty-four laboratories submitted results for the *Campylobacter* panel. The results from these laboratories are summarized below.

<table>
<thead>
<tr>
<th>Number of labs that identified the strain:</th>
<th>QA-070</th>
<th>QA-071</th>
<th>QA-072</th>
<th>QA-073</th>
<th>QA-074</th>
</tr>
</thead>
<tbody>
<tr>
<td>Correctly to the species level</td>
<td>44</td>
<td>12</td>
<td>22</td>
<td>22</td>
<td>22</td>
</tr>
<tr>
<td>Correctly as hippurate negative C. jejuni/coli</td>
<td>NA</td>
<td>NA</td>
<td>15</td>
<td>13</td>
<td>12</td>
</tr>
<tr>
<td>Correctly to the genus level</td>
<td>2</td>
<td>31</td>
<td>11</td>
<td>8</td>
<td>14</td>
</tr>
<tr>
<td>Gram negative rod</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Correctly to the genus or species level but with phenotypic, interpretation, or reporting errors</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Incorrectly identified</td>
<td>3</td>
<td>4</td>
<td>1</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Nonviable</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>50</strong></td>
<td><strong>50</strong></td>
<td><strong>50</strong></td>
<td><strong>50</strong></td>
<td><strong>50</strong></td>
</tr>
</tbody>
</table>

**General Comments**

- Thirty-six labs submitted only phenotypic identification results, which includes traditional phenotypic testing, MALDI-TOF testing, or a combination of traditional and MALDI-TOF testing. Thirteen laboratories submitted a combination of phenotypic and molecular genetic results, with three of those laboratories submitting both MALDI-TOF and traditional phenotypic testing. One laboratory submitted MALDI-TOF results only.
- Forty-eight laboratories correctly reported three or more strains to either the genus or species level based on the testing performed; of those, nine laboratories correctly identified all isolates to the species-level. While the majority of the participants correctly identified or reported the strains to either the species or genus level, phenotypic testing, result interpretation, and report formatting errors were seen. These included:
  - Incorrect results for phenotypic tests including hippurate, indoxyl acetate, oxidase and growth tests. *Campylobacter* spp. can have unique requirements for media and timing for these tests, please contact us if you have any questions about the performance of these tests in your laboratory. See Phenotypic Identification below for more information.
  - General data entry errors occurred in transcribing results from the laboratory worksheet to the reporting form.
  - Contamination or switching of strains with other QA strains or testing control strains.
- Over-interpretation of 16S rRNA gene sequencing data resulting in species-level identifications that were not fully supported by the test results.
- Strains should be reported to the highest level of confidence of correctness and no higher. Disclaimers such as “not jejuni” or “possibly subsp. XXX” should be avoided.

- QA-071 was a urease positive C. lari. MALDI-TOF was able to correctly identify this organism to the species-level, but both PCR and 16S gene sequencing methods cannot always correctly identify this variant as C. lari. If your current testing algorithm cannot distinguish this organism with phenotypic and genetic methods, please feel free to forward isolates to CDC for species identification.

- The public health significance of the Campylobacter species other than C. jejuni and C. coli remains unclear as we have limited species-level surveillance data for other species. MALDI-TOF and molecular genetic testing can aid in the identification of Campylobacter species. If MALDI-TOF or molecular genetic testing is not available, ideally states should have the capacity to identify Campylobacter isolates as either C. jejuni, hippurate negative C. jejuni/ C. coli or Campylobacter species. Laboratories are always welcome to refer non-C. jejuni isolates to the CDC for full identification or confirmation.

**Phenotypic Identification**

- MALDI-TOF-based identification appears to be a useful and accurate tool for Campylobacter species. Twenty-two laboratories reported performing this method. Nineteen laboratories correctly identified all strains tested. Three laboratories were unable to determine a species-level identification for one strain. One laboratory saw an incorrect species-level identification for QA-071 possibly due to strain mix up or contamination. The majority of laboratories used the Bruker MALDI-TOF instrument.

- Common phenotypic testing concerns include:
  - The use of 24 hour growth is recommended when performing phenotypic testing for Campylobacter species identification. In our experience, the use of older growth can lead to test results that are difficult to interpret or erroneous. If the growth at 24 hour is insufficient for testing, the strain should be incubated for an additional 24 hours, subcultured to fresh medium, and grown for an additional 24 hours prior to testing.
  - TSA II agar with sheep blood is not an optimal growth medium for Campylobacter. Growth is generally poor and extended incubation times may be needed to obtain sufficient growth for testing. Brain Heart Infusion (BHI) agar with 5% sheep blood is a more optimal general growth medium for Campylobacter; it is commercially available from Fisher Scientific and Hardy Diagnostic. We are happy to provide the formulation of this medium for laboratories with in-house media preparation capacity.
  - Selective media should not be used for routine culture of pure strains or growth for phenotypic testing because the selective agents can slow or inhibit bacterial growth of some Campylobacter species which may impact the results of phenotypic tests.
  - The hippurate test should be very heavily inoculated (almost milky), incubated at
37°C for two hours, and then developed with ninhydrin for exactly ten minutes using a timer. Longer incubation times can cause overdevelopment of the test, resulting in a false positive. The test can also be over-interpreted. A positive result should be deep purple. A negative result can range from no color change to grey to light purple.

- The H₂S-TSI test should be performed using fresh medium prepared on the day of testing. The use of older medium can lead to unreliable results.
- The nitrate medium routinely used for testing enteric bacteria is made with a peptone basal medium which does not support the growth of all Campylobacter and can lead to false negative results. If using a commercially available nitrate test, ensure it is intended for use with Campylobacter. If preparing the medium in house, use a recipe with heart infusion basal medium as it supports the growth of all Campylobacter species tested to date.
- Naladixic acid and cephalothin susceptibility testing must not be used for species level identification of Campylobacter because it does not produce reliable results due to acquired antimicrobial resistance. If your laboratory is still performing these tests for identification purposes, please discontinue this testing.
- When performing growth and tolerance tests for Campylobacter species, ensure that the tests are performed correctly. Appropriate positive and negative control strains should be included in the tests. A 1 McFarland density bacterial suspension should be used for growth and tolerance tests requiring inoculation with suspensions. Tests should be incubated in the appropriate test conditions for 72 hours prior to reading.
- There are several formulations of MacConkey agar. MacConkey II is the correct formulation for testing Campylobacter.
- If a phenotypic test result is not consistent with the species identified (e.g. aerobic or 25°C growth for C. jejuni), the atypical result should be reviewed for data transcription errors and, if necessary, the test should be repeated to confirm the result. If repeat testing confirms an atypical result, your laboratory may contact us for guidance.

**Molecular Genetic Identification**

- Molecular genetic-based methods are useful tools for identification of the common Campylobacter species. Eight laboratories reported performing PCR testing only. Three laboratories reported performing partial 16S rRNA gene sequencing and PCR testing. Three laboratories reported performing full length or partial 16S rRNA gene sequencing only.
- Common molecular genetic testing concerns include:
  - Full length or partial 16S rRNA gene sequencing cannot reliably differentiate C. jejuni, C. coli and C. lari as the gene sequences from these species can be identical. C. lari can be differentiated from C. jejuni and C. coli using phenotypic methods such as MALDI-TOF or the indoxyl acetate test; C. lari is negative in the indoxyl acetate test whereas C. jejuni and C. coli are positive. Hippurate negative C. jejuni and C. coli strains can be differentiated with PCR assays using gene targets other than the 16S rRNA gene or by MALDI-TOF testing.
It is important to note that partial 16S rRNA gene sequencing cannot differentiate between closely related species such as *C. upsaliensis* and *C. helveticus* or *C. fetus* and *C. hyointestinalis*. The *Campylobacter* 16S rRNA gene contains four variable regions; only two of these variable regions are examined using partial 16S rRNA gene sequencing.

A commercially available PCR assay failed to identify QA-071. This strain is a urease positive *C. lari*, which may not react in PCR assays developed using *C. lari* subsp. *lari* strains.

### 2018 *Escherichia/Shigella* QA Program Summary

Fifty-eight laboratories submitted results for the *Escherichia/Shigella* panel. The results from these laboratories are summarized below.

<table>
<thead>
<tr>
<th>Number of labs correctly determining:</th>
<th>QA-075</th>
<th>QA-076</th>
<th>QA-077</th>
<th>QA-078</th>
<th>QA-079</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genus and species</td>
<td><em>Escherichia coli</em></td>
<td><em>Escherichia coli</em></td>
<td><em>Escherichia vulneris</em></td>
<td><em>Shigella boydii</em></td>
<td><em>Shigella flexneri</em></td>
</tr>
<tr>
<td>O antigen</td>
<td>O157:H7</td>
<td>O103:H2</td>
<td>stx2, eae, ehxA</td>
<td>stx1, eae, ehxA</td>
<td></td>
</tr>
<tr>
<td>O:H serotype (optional)</td>
<td>57</td>
<td>58</td>
<td>NA</td>
<td>51</td>
<td>53</td>
</tr>
<tr>
<td>Stx/Stx (optional)</td>
<td>47</td>
<td>56</td>
<td>NA</td>
<td>31</td>
<td>30</td>
</tr>
<tr>
<td>Stx/Stx type (optional)</td>
<td>33</td>
<td>43</td>
<td>NA</td>
<td>24</td>
<td>24</td>
</tr>
<tr>
<td>Presence of STEC virulence gene eae (optional)</td>
<td>10</td>
<td>11</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Presence of STEC virulence gene ehxA (optional)</td>
<td>6</td>
<td>5</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Presence of invasion marker ipaH (optional)</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>Total</td>
<td>58</td>
<td>58</td>
<td>NA</td>
<td>57</td>
<td>57</td>
</tr>
</tbody>
</table>

NA = not applicable
General Comments

- Seven laboratories (12%) correctly identified and characterized all four strains with respect to genus/species, O antigen and Shiga toxin type/genotype for STEC, and genus/species (subgroup) and serotype for Shigella.
- Whole genome sequencing technology is advancing rapidly, and its implementation will allow public health laboratories to identify bacteria from their genome sequences and extract a variety of subtype information from these data (O:H serotype, virulence and antimicrobial resistance genes as well as a variety of sequence-based subtypes). Until this technology is fully implemented, we encourage laboratories to phenotypically determine the O antigens of the top seven Shiga toxin-producing Escherichia coli (O26, O45, O103, O111, O121, O145 and O157) and the type of Shiga toxin(s) produced (Stx1 and Stx2) or the genes encoding these toxins (stx1 and stx2) and O antigens. We also encourage states to serotype Shigella isolates (determine the O antigen) because shigellosis is a nationally notifiable disease for which serotype information is collected through the Laboratory-based Enteric Disease Surveillance (LEDS) system. Serotype results are helpful in monitoring trends and detecting clusters of infection due to serotypes other than S. sonnei. If resources permit, please consider adding this testing to your enteric work-up.

QA-075
- This isolate is a typical representative of Shiga toxin-producing E. coli O157:H7.

QA-076
- This isolate is a typical representative of Shiga toxin-producing E. coli O103:H2.

QA-077
- Escherichia vulneris was included to assess your methods for identifying Escherichia species. Regrettably, we believe some vials of this strain may have been contaminated with another Escherichia species. For this reason, QA-77 is ungraded on the panel.

QA-078
- This isolate is a typical representative of Shigella boydii 4.

QA-079
- For many years, Shigella dysenteriae type 1 was the only serotype recognized to produce Shiga toxin; however, over the past several years, sporadic and outbreak-associated infections due to Shiga toxin-positive Shigella serotypes, including Shigella sonnei, S. dysenteriae types 4, S. flexneri types 2a and Y variants, and Shigella boydii types 1 and 19, have been described (1-6). These infections have been associated with travel to Haiti, the Dominican Republic, French Guiana, Mexico, Morocco and Ukraine. Most isolates described to date are positive for the stx1; however, a S. sonnei isolate positive for stx2 was reported in one study (6). The prevalence of Shiga toxin-positive shigellae in the US is currently not known, but it should be possible to readily assess this prevalence once routine WGS testing is implemented for Shigella. When attempting to isolate a Shiga toxin-positive organism, it is worth keeping in mind that the Shiga toxin genes may reside within Shigella.


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**2018 Salmonella QA Program Summary**

Sixty laboratories submitted results for the *Salmonella* panel. Fifty-five labs performed full serotyping; those results are summarized below.

<table>
<thead>
<tr>
<th>Number of labs identifying the strain as:</th>
<th>QA-080</th>
<th>QA-081</th>
<th>QA-082</th>
<th>QA-083</th>
<th>QA-084</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abaetetuba I 11:k:1,5</td>
<td>50</td>
<td>23</td>
<td>51</td>
<td>39</td>
<td>46</td>
</tr>
<tr>
<td>Orion I 3,15:y:1,5</td>
<td></td>
<td>48:z35:-</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chicago I 28:r,i:1,5</td>
<td></td>
<td></td>
<td>13:15:y:1,5</td>
<td>9</td>
<td>2</td>
</tr>
<tr>
<td>Enteritidis I 9,12:g,m:-</td>
<td></td>
<td></td>
<td></td>
<td>19,12:g,m:-</td>
<td></td>
</tr>
<tr>
<td>the expected serotype</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>a serotype other than the expected one</td>
<td>1</td>
<td>17</td>
<td>1</td>
<td>9</td>
<td>2</td>
</tr>
<tr>
<td>a monophasic or rough variant</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>a partial serotype</td>
<td>3</td>
<td>15</td>
<td>2</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>Total</td>
<td>55</td>
<td>55</td>
<td>55</td>
<td>55</td>
<td>55</td>
</tr>
<tr>
<td>% expected serotype</td>
<td>91%</td>
<td>42%</td>
<td>93%</td>
<td>69%</td>
<td>84%</td>
</tr>
</tbody>
</table>

**General comments:**

- Twenty-six labs performed only conventional serotyping, 7 labs performed only molecular serotyping, 9 labs performed both full conventional and molecular serotyping, and 13 labs performed molecular serotyping supplemented with conventional serotyping when needed. Seven labs used whole genome sequencing to perform serotyping.
• Overall, 19 labs (35%) correctly serotyped all five strains. Among the 275 serotype determinations in the 55 labs, 209 (76%) were the expected serotype; 19 (7%) were a serotype other than the expected one; three (1%) were monophasic variants; and, 44 (16%) were partially or not identified.

• Many of the partially serotyped strains were due to lack of appropriate antisera. While CDC is running out of some specificities of Salmonella antisera, all the reagents necessary to serotype the strains in this panel are available from CDC. Please contact us at salmonella@cdc.gov for instructions on how to request Salmonella serotyping reagents from CDC.

QA-081

• Including the additional 5 labs that only performed serogrouping or identification, 24 (40%) labs identified this strain as either Salmonella bongori or Salmonella bongori subspecies V, 7 (12%) as Salmonella enterica subspecies bongori, 8 (13%) as Salmonella enterica subspecies arizonae, 7 (12%) as Salmonella enterica subspecies enterica, 2 (3%) as Salmonella enterica subspecies salamae, 1 (2%) as Salmonella enterica subspecies diarizonae, and 11 (15%) labs did not report identification to the species or subspecies level for this strain.

• For clarity of distinguishing Salmonella bongori as a separate species as opposed to a former subspecies of Salmonella enterica, we choose to no longer report Salmonella bongori serotypes with a Roman numeral “V”. We believe that an antigenic formula without a roman numeral implies a Salmonella bongori serotype. These serotypes are, however, still defined in the Kauffmann-White Scheme using the Roman numeral “V”.

QA-082

• We included this strain in the panel to determine whether labs are testing for ancillary O antigens. Ancillary O antigens will be more difficult to detect when we transition to WGS; but, they are still important for differentiating between some recognized serotypes, e.g., serogroups O:3,10 vs O:1,3,19.

• Twenty-eight labs tested for and reported O:15 (51%), 7 (13%) labs tested for and got a negative result for O:15, and 20 (36%) labs did not test for O:15.

QA-083

• The antigenic formula as defined in the Kauffmann-White Scheme for serotype Chicago is I 28:r,[i]:1,5, indicating variable presence of H:i. While the serotype designation does not change with the presence or absence of H:i, routine confirmation of these variable antigens may improve our understanding of Salmonella diversity.

• Twenty-seven (49%) labs tested for both H:r and H:i, 25 (46%) labs only tested for H:r, and 3 (5%) labs only tested H:i.

QA-084

• This is a very atypical strain of serotype Enteritidis that is both Vi positive and sdf-negative via genetic methods.

• Serotype Gallinarum, a bird pathogen that should be very rare in the US. It is nonmotile but has a non-expressed H:g,m fliC allele making its antigenic profile the same as
serotype Enteritidis when determined by genetic methods. The $sdf$ gene is used in the Luminex SSA to differentiate common Enteritidis strains ($sdf+$) from serotype Gallinarum ($sdf-$). There are, however, several lineages of $sdf-$ Enteritidis circulating that are relatively rare based on surveillance data. These strains are highly divergent from both the common $sdf+$ Enteritidis lineage and from Gallinarum. For additional information, please refer to the following study:
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4178404/.

- Vi, a marker represented by viaB in the Luminex SSA, is characteristic of serotypes Typhi, Paratyphi C, and Dublin based on the Kauffmann-White Scheme, but may occur sporadically in other serotypes. The presence/absence of Vi should only be considered for those serotypes listed above and disregarded for all other serotypes.

- We are currently working to incorporate a marker specific to serotype Gallinarum in the SeqSero database that will allow for clear distinction from $sdf-$ Enteritidis lineages via WGS analysis.

Nomenclature
- The use of non-standard antigenic formulas to represent Salmonella serotypes continues to be a problem. We understand that including all relevant antigens in the formula is not necessary for those that understand Salmonella serotyping. Given the complicated nature of Salmonella serotype designations and to try to improve understanding, we think it is important to use clear, correct, consistent nomenclature according to the White-Kauffmann-Le Minor Scheme.
  - For H antigen complexes, the primary antigen should be listed, e.g., “1,5” not “5.”
  - Antigenic factors within an antigen designation are separated by commas, e.g., g,s,t and e,n,x.
  - Generic antigenic formulas for serotypes may contain antigens that are underlined, in square brackets ([]), in curly brackets ({ }), or in parentheses; they denote antigenic variability within a serotype. For a strain that has been serotyped, list only the antigens that were tested for and detected in the antigenic formula. E.g., the antigenic formula for serotype Typhimurium is I 4,[5],12:i:1,2; the antigenic formula for a strain of serotype Typhimurium is I 4,5,12:i:1,2 or I 4,12:i:1,2.

Please contact us at salmonella@cdc.gov with any questions regarding these results or for information on ordering Salmonella antisera.

2018 Other Enterobacteriaceae QA Program Summary

Fifty three laboratories submitted results for the ‘Other Enterobacteriaceae’ panel. Six laboratories (11%) correctly identified both isolates to species. Of these, five laboratories used DNA sequencing methods (either 16S rRNA gene or whole genome sequencing) to derive the correct identification and one laboratory used MALDI TOF (platform not reported). A total of 20 laboratories used MALDI TOF which in general provided a reliable identification to genus but not to species.
QA-085
This strain is Cronobacter sakazakii. A total of 28 laboratories correctly reported Cronobacter sakazakii, and 17 additional laboratories correctly identified the isolate as Cronobacter sp. or C. sakazakii complex. One laboratory reported C. sakazakii/C. malonaticus and one laboratory correctly ruled out more common enteric pathogens. Six laboratories reported other Enterobacteriaceae with Enterobacter being the most common genus reported.

QA-086
This strain is Cronobacter turicensis. Six laboratories (11%) correctly identified the isolate as Cronobacter turicensis. A total of 27 laboratories correctly identified the isolate as Cronobacter sp. or Cronobacter sakazakii complex. One laboratory correctly ruled out more common enteric pathogens. Fourteen laboratories correctly placed QA-085 into the genus Cronobacter but misidentified the species (12, C. sakazakii; 1, C. malonaticus; 1, C. universalis). One laboratory reported Enterobacter cloacae and one reported Enterobacter sp. Two laboratories reported Enterobacter sakazakii. It is important to note that E. sakazakii equates to Cronobacter sakazakii complex, as the taxonomic revision of E. sakazakii split the single species into multiple species of Cronobacter. Thus, an identification of E. sakazakii with commercial panels should be reported as “Cronobacter sp.” or “Cronobacter sakazakii complex.” Cronobacter can be differentiated from Enterobacter by its production of yellow pigment on TSA. Specific phenotypic tests could augment results from commercial panels to identify QA-085 and QA-086. Cronobacter turicensis produces acid upon fermentation of dulcitol, whereas C. sakazakii does not (nor does its close relative, C. malonaticus). In addition C. turicensis (and C. malonaticus) typically utilizes malonate but C. sakazakii does not. Note that one group of C. sakazakii does utilize malonate, and other phenotypic characteristics can also be variable and thus phenotypic identification can be unreliable. MALDI TOF can identify isolates to the genus level but may not distinguish among all the Cronobacter species. Differentiating C. sakazakii and its closest relative C. malonaticus is especially problematic. These two species cannot reliably be differentiated using 16S rRNA gene sequencing, but gene trees constructed from other housekeeping gene sequences clearly group Cronobacter isolates into the seven distinct species. Gene sequencing (or genome sequencing) and phylogenetic analysis is the most reliable approach for identifying Cronobacter species.

Cronobacter sakazakii infections are rare but life-threatening, particularly in neonates, where it can cause potentially fatal septicemia and meningitis. Powdered infant formula (PIF) has been shown to be a source of illness and outbreaks of C. sakazakii among neonates, but unopened containers of PIF have not been implicated in any recent cases of neonatal illness investigated by the CDC. Any Cronobacter species in PIF is considered an adulterant according to the Code of Federal Regulations. Although Cronobacter sakazakii is of greatest public health concern, Cronobacter malonaticus and Cronobacter turicensis also cause disease in humans, and C. turicensis has been associated with neonatal illness. The isolation of any Cronobacter from a neonate should be reported to the CDC for case investigation and the isolate forwarded for species identification and additional testing.
*Yersinia, Cronobacter* and other *Enterobacteriaceae* isolates can be sent to CDC (Unit 16) for identification as needed. The performance characteristics of sequencing a segment of the *rpoB* gene for enteric bacterial identification has been validated and the gene segment is useful for identification of enteric bacteria including many *Enterobacteriaceae*. As the CDC and the network of public health laboratories transition to whole genome sequencing, *Yersinia enterocolitica* and *Cronobacter sakazakii* will be among the organisms for which identification and genome-based multilocus sequencing typing (MLST) will be conducted on a routine basis.