Performance of the ImmunoCard STAT! E. coli O157:H7 Test for Detection of Escherichia coli O157:H7 in Stools

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ImmuzenCard STAT! E. coli O157:H7 (Meridian Diagnostics, Inc., Cincinnati, Ohio) is a novel rapid (10-min) test for the presence of Escherichia coli O157:H7 in stools. The test may be performed either directly on stool specimens or on an overnight broth culture of stool. In a multicenter prospective study, 14 of 14 specimens positive by culture for E. coli O157:H7 were positive by the ImmunoCard STAT! O157:H7 test, and there were no false positives from 263 culture-negative specimens. In a retrospective study, the test was positive in 339 (81%) of 417 stored culture-positive specimens and the specificity was 95% (98 of 103 specimens). No false positives were associated with alternate stool pathogens. The ImmunoCard STAT! O157:H7 test has high sensitivity and specificity.

RESULTS

The results of the clinical study are shown in Table 1. Twelve of 14 culture-positive stool specimens were positive by both tests. One specimen was positive after incubation but negative by the direct test, and this specimen yielded a single colony of E. coli O157:H7 after an exhaustive search, in excess of normal clinical routine. One was positive by the direct test but negative by the incubation test. This specimen also yielded the organism after an augmented search. A total of 14 of 14 stool specimens were positive in at least one of the tests (direct or after incubation). There were no false positives among 263 culture-negative specimens.

Table 2 shows the results of the retrospective study. The test results were positive for 339 (81%) of 417 culture-positive specimens, and the specificity was 95% (98 of 103 specimens). Of the 339 test-positive specimens, 328 had been frozen without preservation and 11 had been preserved in Cary-Blair medium. The day of illness on which the specimen was taken was available for 361 of the 417 culture-positive specimens in the retrospective study. The direct test result was positive for 85% (269 of 317 specimens) of stool samples taken on or before the
fifth day of illness and for 73% (32 of 44 specimens) stool samples taken on the sixth day or later.

**DISCUSSION**

In the clinical study the ICS test result was positive for 14 of 14 culture-positive stools (Table 1). This number suggests a high sensitivity, when considered with the overall 81% sensitivity demonstrated in the retrospective study, which is probably an underestimate (see below and Table 2). In the clinical specimen for which the result was negative by the direct test but positive after incubation, it is likely that initially the number of organisms was too small to be detected and that the incubation step enhanced the population of *E. coli* O157:H7 over the detection threshold. Antibiotic in the specimen might explain the result on the specimen which was positive by the direct test but negative after incubation, although we were not able to verify this. The ICS test requires the presence of both antigens (O157 and H7), and the capture system requires the structural integrity of the organisms, to ensure the comigration of the two antigens along the membrane. We suggest that the organisms were killed and disrupted by incubation with antibiotic, although enough bacteria were present in the fresh specimen to yield a positive direct test result. This requirement for the presence of two separate antigens on intact organisms to yield a positive result is probably also the explanation for the specificity of the test. Both of the apparent false-positive results were resolved by an enhanced search for viable organisms, suggesting that the sensitivity of this test for this patient population is comparable to that of standard culture. The specificity of ICS in the clinical study was 100% (263 of 263 specimens).

The sensitivity of the direct test performed on the stored specimens was 85% for specimens taken on or before the fifth day of illness. The sensitivity of the test is probably dependent on the number of organisms present in the specimen submitted; it is likely that storage conditions had an impact on the number of organisms. Therefore, the overall sensitivity of 81% on frozen and thawed specimens in the retrospective study using stored specimens

<table>
<thead>
<tr>
<th>ICS test result</th>
<th>No. of specimens whose result by culture was:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Positive</td>
</tr>
<tr>
<td></td>
<td>No CB&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Positive (no. of specimens)</td>
<td>328</td>
</tr>
<tr>
<td>Negative (no. of specimens)</td>
<td>74</td>
</tr>
<tr>
<td>Sensitivity (%)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>82</td>
</tr>
<tr>
<td>Specificity (%)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>NA</td>
</tr>
</tbody>
</table>

<sup>a</sup> Specimens frozen without transport medium (Cary-Blair [CB] medium).
<sup>b</sup> Specimens preserved in Cary-Blair (CB) medium.
<sup>c</sup> *E. coli* O157:H7 culture.
<sup>d</sup> No significant difference between values for culture-positive specimens (*P* > 0.05, *χ²* test).
<sup>e</sup> NA, not applicable.
study would underestimate the sensitivity of the direct test in clinical use. It has been demonstrated in cases of VTEC diarrhea that the number of organisms in the stool decreases progressively after the first few days of infection (8). This may be the reason for the lower sensitivity of the test in specimens taken after the fifth day of the illness. The number of specimens in Cary-Blair medium was not large enough to demonstrate a total absence of effect due to this medium, but the numbers in Table 2 do demonstrate that any effect due to Cary-Blair medium is not great. The absence of false-positive results on the 50 specimens from which alternate pathogens were isolated is reassuring, particularly in the case of Campylobacter sp. infections which, like VTEC, frequently present with bloody diarrhea.

Commercially available rapid tests for VTEC include those which detect verotoxin in stools or culture supernatants and those which detect structural bacterial antigen (4, 5, 7, 9). ICS is more sensitive and specific than the Premier E. coli O157 test previously reported (7).

The ICS test is simple to perform, the direct test gives a result within 10 min, and the test will be of particular value in areas where E. coli O157:H7 is the predominant VTEC serotype. Early detection of patients at risk will permit closer observation for signs of the onset of hemolytic-uremic syndrome, and rapid laboratory diagnosis also facilitates management of outbreaks.

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REFERENCES