Detection of non-jejuni/coli Campylobacter species from stool with an immunochromatographic antigen detection assay

Brianne A. Couturier, Marc R. Couturier, Kimberly J. Kalp, and Mark A. Fisher

ARUP Laboratories
An enterprise of the University of Utah and its Department of Pathology

Abstract

Background: Campylobacter spp. are difficult to recover in culture due to sub-optimal specimen transport and/or storage conditions and lack of universal culture procedures. However, Campylobacter antigens may persist in clinical specimens in the absence of viable organisms. The variable clinical specificity reported for commercial Campylobacter antigen detection assays remains controversial. We sought to resolve the inconsistency observed in several studies regarding the clinical specificity of Campylobacter antigen detection by evaluating an immunochromatographic assay for Campylobacter antigen in comparison to micro-aerobic culture, with discrepant specimens interrogated by PCR. In addition, we examined the stability and reactivity profile of other Campylobacter spp. with the antigen assay.

Methods

- **Materials**: 500 fecal specimens submitted over a 4-month period for routine stool cultures or Campylobacter-specific cultures were tested with the Immunocard® Campy agar assay (Meridian Bioscience, Cincinnati, OH). Campylobacter spp. were identified by standard biochemicals and MALDI-TOF. Direct stool PCR was performed on antigen- or culture-positive stools. PCR primers targeted a Campylobacter-specific region of the 16S rRNA gene for genus confirmation and species-specific primers for Campylobacter upsaliensis.

- **Results**: Of the 500 specimens analyzed, five (1%) samples were culture-positive for Campylobacter jejuni and fifteen (3%) were Campylobacter non-jejuni/non-coli positive. Four specimens were both culture and antigen-positive. Ten antigen-positive specimens were PCR-positive for Campylobacter spp., including three that were PCR-positive for C. upsaliensis. Tissue culture and antigen-positive stool specimens were tested for stability, and remained positive for both assays, for at least 5 days at 4°C.

- **Conclusion**: Campylobacter-specific antigen reactivity was detected in several culture-negative/PCR-positive specimens. C. upsaliensis, a pathogenic species that is traditionally difficult to recover in routine stool cultures, was detected in those of these culture-negative specimens. This study provides evidence that antigen testing may expand the detection of campylobacters.

Table 1: Culture results versus antigen detection assay

<table>
<thead>
<tr>
<th>Culture Results</th>
<th>POS</th>
<th>NEG</th>
<th>Grand Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>POSSITIVE</td>
<td>4</td>
<td>1</td>
<td>5</td>
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<tr>
<td>NEGATIVE</td>
<td>11</td>
<td>484</td>
<td>495</td>
</tr>
<tr>
<td>Grand Total</td>
<td>15</td>
<td>485</td>
<td>500</td>
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</tbody>
</table>

A sensitivity of 80% and specificity of 98% relative to the gold standard of culture was calculated.

Table 2: Direct Campylobacter spp. PCR and sequencing results for specimens generating discrepant results between antigen detection and culture

<table>
<thead>
<tr>
<th>Study ID</th>
<th>C. jejuni</th>
<th>C. concisus</th>
<th>C. upsaliensis</th>
<th>C. lari</th>
<th>C. lari</th>
<th>C. upsaliensis</th>
<th>C. upsaliensis</th>
<th>C. upsaliensis</th>
<th>C. upsaliensis</th>
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<tr>
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<td>NEGATIVE</td>
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Table 3: A comparison of antigenic cross-reactivity for multiple clinical or reference Campylobacter isolates

<table>
<thead>
<tr>
<th>Campylobacter spp.</th>
<th>STAT Campy Result</th>
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<tbody>
<tr>
<td>C. coli (n=1)</td>
<td>POSITIVE</td>
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<tr>
<td>C. curvus (n=2)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. concisus (n=1)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. fetus (n=1)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. gracilis (n=1)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. jejuni (n=1)</td>
<td>POSITIVE</td>
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<tr>
<td>C. lari (n=2)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. rectus (n=1)</td>
<td>NEGATIVE</td>
</tr>
<tr>
<td>C. sputorum (n=1)</td>
<td>NEGATIVE</td>
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<tr>
<td>C. upsaliensis (n=7)</td>
<td>POSITIVE</td>
</tr>
<tr>
<td>C. vulgaris (n=1)</td>
<td>NEGATIVE</td>
</tr>
</tbody>
</table>

Table 4: Relative sensitivity of antigen detection from C. jejuni and C. upsaliensis cultures

<table>
<thead>
<tr>
<th>McFarland Standard</th>
<th>C. jejuni</th>
<th>C. upsaliensis</th>
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<tbody>
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<td>2.0</td>
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<td>1:10 dilution</td>
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<td>1:100 dilution</td>
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Acknowledgements

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References