Comparison of VIDAS® GDH automated immunoassay with Cepheid GeneXpert® C. difficile PCR assay and an in-house PCR assay for GluD, for the detection of C. difficile in faecal samples

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Introduction

Laboratory diagnosis of Clostridium difficile infection (CDI) has traditionally involved the detection of organism-specific toxins. For cell-culture specific or enzyme immunoassays. Recently the poor prognostic value of these assays has been highlighted and alternative markers of CDI have been sought. Glutamate dehydrogenase (GluD) is a cell-surface associated enzyme found in many bacteria. C. difficile-specific GDH has been shown to be highly conserved between different PCR-changes of C. difficile. Assays targeting C. difficile-specific GDH have been developed and a recent meta-analysis showed that GDH had a sensitivity and specificity of 90% when compared to culture. GDH cannot determine the toxicogenic status of C. difficile however. It must be used as part of an algorithm for CDI detection, most commonly alongside toxin detection or polymerase chain reaction (PCR) for toxin genes. Although initial algorithms of GDH followed by either cell-culture or cytotoxicity assay or cell-culture toxin assay on stool enriched cultures improved sensitivity, compared with standalone toxin enzyme immunoassays (EIA), the turnaround time could be up to 3 days. Using a combination for the second step improves the specificity of two-step algorithms but are limited by the sub-optimal sensitivity of currently available toxins EIA. Whilst there have been many algorithmic approaches, a recent review led by the Department of Health in England advising using a two-stage algorithm, with GDH or toxins (PCR) as the initial screen, for detection of CDI.

Objectives:

To determine a new automated immunoassay, VIDAS® GDH, with the Cepheid GeneXpert® C. difficile toxigenic assay by Polymerase Chain Reaction (PCR) assay and an in-house PCR assay for GluD (Gdh) of C. difficile, for the laboratory diagnosis of CDI.

Methods

One hundred and sixty seven cystitis-positive diarrhoeal faecal samples submitted to Leeds Teaching Hospitals NHS Trust, UK, were submitted for testing from June 2010 to August 2011 (n=167) as part of 3, 7, and 9 tests sites in Leeds, Berlin and St Etienne. All samples were diarrhoea (took the shape of the container), were <5 days old and all had a positive PCR sample characterisation to culture. The VIDAS® GDH assay was performed on a VIDAS instrument (bioMerièux, France), whilst the GeneXpert® C. difficile assay was performed on a smart cycler (Cepheid, France). Both laboratories used the same C. difficile and C. difficile PCR assay. All samples were diarrhoea and tested using a CE marked GDH assay (Alere, USA) and cultured directly onto C. difficile ID® agar (bioMerièux). GDH was performed on a VIDAS instrument (bioMerièux, France), whilst the GeneXpert® C. difficile assay was performed on a smart cycler (Cepheid, France). Both laboratories used the same C. difficile and C. difficile PCR assay. All samples were diarrhoea and tested using a CE marked GDH assay (Alere, USA) and cultured directly onto C. difficile ID® agar (bioMerièux). GDH was performed on a VIDAS instrument (bioMerièux, France), whilst the GeneXpert® C. difficile assay was performed on a smart cycler (Cepheid, France).

Results

Table 2. Sensitivity, specificity and positive and negative predictive values and Pearson’s correlation of the VIDAS GDH assay compared with two different PCR assays.

<table>
<thead>
<tr>
<th>Test</th>
<th>Sensitivity 95% CI</th>
<th>Specificity 95% CI</th>
<th>PPV 95% CI</th>
<th>NPV 95% CI</th>
<th>Pearson’s correlation</th>
</tr>
</thead>
<tbody>
<tr>
<td>VIDAS GDH</td>
<td>92.6</td>
<td>91.0</td>
<td>0.87</td>
<td>0.97</td>
<td>0.92</td>
</tr>
<tr>
<td>VIDAS GDH after dissociation</td>
<td>50.5</td>
<td>69.3</td>
<td>0.39</td>
<td>0.77</td>
<td>0.86</td>
</tr>
</tbody>
</table>

Table 3. VIDAS GDH false-negative sample characterization

Table 4. VIDAS GDH false-positive sample characterization

Discussion

The VIDAS GDH assay has been compared here with two PCR assays (toxin gene detection & GDH gene detection). It showed a sensitivity and specificity of 92.6% and 91.0% respectively, compared with PCR for GDH. The VIDAS GDH assay had a sensitivity and specificity of 90% and 90% respectively, compared with Cepheid GDH PCR. The VIDAS GDH assay was used in a comparison to examine the VIDAS GDH assays ability to detect GDH from a sample, not diagnose CDI. The comparison with the Cepheid GDH PCR assay demonstrates that, if expected, the specificity decreased to 88.1%. This result indicates that the VIDAS GDH assay showed a higher positive predictive value (PPV) (92.6%) than the Cepheid GDH PCR assay (90.7%). The VIDAS GDH assay showed a high negative predictive value (NPV) as compared to culture and direct cytotoxic culture, which correlates with the high NPV seen with other GDH assays. Indeed, the high sensitivity of this methods has lead to the inclusion of GDH as a first (screening) assay of two-stage algorithms for CDI diagnosis, although there is little consensus on the best algorithm.

Conclusions

The VIDAS GDH assay has comparable accuracy to the GeneXpert® C. difficile PCR assay and our in-house GluD PCR assay.

The VIDAS GDH assay could be an option as a first-line (screening) test in a two-step C. difficile testing algorithm.

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References