Development of an EQA Scheme for Norovirus Testing

Hannah McGregor [1] [2], Dr Brigitte Senechal [2], Dr Vivienne James [2]
1 University of Surrey, 2 UK NEQAS for Microbiology

Aims
• To determine the levels of interest in an EQA scheme for Norovirus testing.
• To determine testing practice for Norovirus detection.
• To prepare a pre-pilot distribution of stool samples for the detection of Norovirus and analyse the results reported.

Introduction
Norovirus is the most common cause of gastroenteritis [1]. It is particularly prevalent in enclosed environments such as hospitals, cruise ships and nursing homes. The virus has a low infectious dose and is spread through the faecal-oral route. Outbreaks of the virus in hospitals cost the NHS an estimated £13 million a year due to ward closures and staff absences. [2] Rapid identification is important for the implementation of infection control measures. Identification assays are front line tools and the results will influence outbreak investigation. To assess the quality of such assays, UK NEQAS intends to develop an EQA scheme beginning with a pre-pilot distribution for Norovirus identification.

Questionnaire

In March 2011, a questionnaire was sent to all existing UK NEQAS for Microbiology participants asking: if they test for enteric viruses (Norovirus, Rotavirus and Adenovirus 40, 41), which assays are used and if the participant would be interested in an EQA scheme provided by UK NEQAS.

Method
Out of 243 replies, 161 (66%) laboratories routinely test for at least one enteric virus. (94 UK laboratories).

97 (60%) routinely perform a Norovirus detection assay. (44 UK laboratories).

77 (79%) would be interested in participating in a UK NEQAS EQA scheme for Norovirus, including 38 UK laboratories (86%).

Pre-pilot distribution

Table 1 – The six specimens sent to participants

<table>
<thead>
<tr>
<th>Specimen number</th>
<th>RT-PCR</th>
<th>CT value</th>
<th>Genotype</th>
<th>EIA</th>
<th>Overall result</th>
</tr>
</thead>
<tbody>
<tr>
<td>0829</td>
<td>Pos</td>
<td>24.4</td>
<td>GII</td>
<td>Positive</td>
<td>RNA +ve Ag +ve</td>
</tr>
<tr>
<td>0829</td>
<td>Pos</td>
<td>20.0</td>
<td>GII</td>
<td>Positive</td>
<td>RNA +ve Ag +ve</td>
</tr>
<tr>
<td>0829</td>
<td>Pos</td>
<td>13.6</td>
<td>GII</td>
<td>Negative</td>
<td>RNA +ve Ag +ve</td>
</tr>
<tr>
<td>0829</td>
<td>Neg</td>
<td>-</td>
<td>GII</td>
<td>Negative</td>
<td>RNA -ve Ag -ve</td>
</tr>
<tr>
<td>0829</td>
<td>Pos</td>
<td>20.9</td>
<td>GII</td>
<td>Negative</td>
<td>RNA -ve Ag +ve</td>
</tr>
<tr>
<td>0829</td>
<td>Pos</td>
<td>17.9</td>
<td>GII</td>
<td>Positive</td>
<td>RNA +ve Ag +ve</td>
</tr>
</tbody>
</table>

Across all six specimens, the most sensitive method appears to be the molecular method. The enzyme Immunoassays (EIAs) detected only the strongest positive specimens (0828, 0829 & 0833). The rapid immunoassay appears to be the least sensitive detection method (Fig 4).

Figure 4 – Percentage of correct results (by method)

Genotyping was carried out by nine laboratories; all of which stated the correct genogroup for the GII specimens. One laboratory genogrouped the negative specimen.

Conclusions
• A larger volume of sample may need to be distributed in the future if the sample is not to be further diluted.
• Dilution of faecal specimens to increase the volume is not ideal as diluted specimens may not be suitable for the less sensitive assays that detect Norovirus antigen. Pooling the same genogroup Norovirus could be considered for future distributions.
• Norovirus antigen is not as stable as the RNA. Faecal material for specimens needs to be as fresh as possible and may require an antigen stabiliser.
• A stability study is currently on-going; testing for the presence of the antigen in specimens stored at different temperatures, with or without adding a stabiliser. This is to establish the best conditions for faecal material collection and for specimen storage.

Acknowledgements
Brigitte Senechal, (Scheme Manager), Vivienne James, (Director of UK NEQAS), UK NEQAS staff, Enteric Virus Unit, Colindale HPA and the Participating Laboratories.

References