REGIONAL & NATIONAL NETWORK FOR THE DETECTION & CHARACTERISATION OF NOROVIRUSES

Enteric Virus Unit, Virus Reference Department
Centre for Infections, HPA, Colindale, London

Newsletter Date:
December 2009 – January 2010

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SUMMARY

- Between December 2009 – January 2010, 175 norovirus strains were genotyped from outbreaks occurring across England
- In total, 7 different norovirus genotypes were detected during this period
- 85% of outbreaks were associated with GII noroviruses
- Norovirus GII-4 variant strain 4 (GII-4v4) was the most frequently detected variant strain

INTRODUCTION

Noroviruses are a major aetiological agent of outbreaks of gastroenteritis, and as such it is important to maintain surveillance of the molecular epidemiology of these viruses. Strain surveillance carried out in the Enteric Virus Unit at CfI in Colindale, involves the characterisation of the first 20 norovirus positive outbreaks occurring in each region in England at the beginning, middle and end of the norovirus season (September–October, December–January and March–April).

QUALITY ASSURANCE DATA

The 2009/2010 quality assessment panel was distributed to 10 HPA laboratories, and a further 7 laboratories across England, Wales and Northern Ireland also requested the panel. The participants in the 2009/2010 quality assessment panel

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</table>

% Correct: 100 88 100 100 100 100 100 100 100 100 100 100 100 100 100 100 100 100 100 100

The panel was comprised of 8 faecal samples: two samples positive for GI norovirus, four samples positive for GII noroviruses, and two norovirus negative specimens. The results returned from participating laboratories are surmised in the table below. Results discrepant from those found in EVU are shown in blue.

Ten laboratories had a 100% success rate, and all participating laboratories returned results ≥75% correct. Seventeen laboratories provided qRT-PCR data (Ct values in the table below), and the standard deviation of the Ct values was between 2.9-4.2, however Ct values within samples varied by >3 cycles.
During the period December 2009 – January 2010, a total of 213 samples from norovirus-positive outbreaks of gastroenteritis from around England were received for genotyping. The target number of strains was received from the East of England, North West, South West, South East Yorkshire and Humberside, but not form London, the North East or the Midlands. In total, 175 (82%) of these were genotyped, and 7 genotypes were detected. The typing data is surmised in the table. Viruses belonging to the GII-4 genotype were most prevalent, being associated with 85% of typed outbreaks.

<table>
<thead>
<tr>
<th>Region</th>
<th>GI-4</th>
<th>GI-6</th>
<th>GII-1</th>
<th>GII-3</th>
<th>GII-4</th>
<th>GII-6</th>
<th>GII-7</th>
<th>Total Genotyped Outbreaks</th>
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<td>Yorkshire &amp; Humberside</td>
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<td>149</td>
<td>10</td>
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</tbody>
</table>

Norovirus Outbreak Settings Data

From the samples received during December 2009 and January 2010, norovirus-positive outbreaks were mainly associated with 3 settings across England (Figure 2): hospitals and nursing homes; schools, nurseries and childcare centres; and, restaurant & foodborne. The norovirus genotypes associated with each of these settings were analysed (Figure 3). All seven of the genotypes detected during this period were associated with outbreaks in hospitals and nursing homes (Figure 3).
**GII-4 NOROVIRUS DIVERSITY**

The most frequently detected norovirus genotype was GII-4, consistent with previous years. As a measure of genetic diversity in the circulating virus population, the GII-4 norovirus strains were further characterised to determine the GII-4 variant type, as described in Gallimore et al. (2007) (Figure 4).

Multiple variants of GII-4 strains were found co-circulating, 6 in total. Variant type 4 (GII-4v4) was the most frequently strain, followed by GII-4v6 then GII-4v10 (Figure 4). The South West region showed the most diversity, with five of the six variant strains detected in this region (Figure 4).

The distribution of GII-4 variants according to setting is shown in Figure 5.

The emergence of novel GII-4 norovirus variant strains has previously been associated with epidemic waves of gastroenteritis (Lopman et al., 2004). Surveillance of strain diversity therefore includes the study of changes within GII-4 strains which may lead to the emergence of an antibody escape mutant strain.

As previously described (Allen et al., 2008), the hypervariable P2-region of the ORF2 (VP1/capsid) gene can be sequenced in order to determine how genetic changes may lead to alterations in the three-dimensional shape of the capsid protein, which ultimately may result in a virus able to evade existing immunity (Allen et al., 2009). This is similar to the evolutionary process described for influenza A viruses, and known as epochal evolution or the neutral network model of evolution.

Analysis of the hypervariable P2 region of the GII-4 variant strains detected between December 2009 – January 2010 showed that the dominant variant differs in the three-dimensional structure of the previously identified variant-specific epitopes from the epidemic strains found in 2002 and 2006. Interestingly, this variant strains emerged in 2007 and gradually displaced the 2006 epidemic stain, becoming predominant since 2009. The detailed analysis of the P2 domain is also being used as a tool for tracking outbreaks and monitoring transmission events within and between hospitals and food-handler related outbreaks (Xerry et al 2008; 2009).
SURVEILLANCE OF HOSPITAL OUTBREAKS

Since January 2009 the HPA in collaboration with the Infection Prevention Society set up a dedicated system for recording outbreaks of norovirus occurring in hospitals in England. Infection control staff enter data on outbreaks as they occur using a secure web-enabled reporting form. This form, along with data and the current report on hospital norovirus outbreak reporting is available at http://www.hpa-bioinformatics.org.uk/noroOBK/.

In the first year, 713 outbreaks were reported on the database, with 71% of outbreaks having laboratory confirmation of norovirus. We are interested in developing this surveillance further by matching outbreaks entered into the database with samples received for strain surveillance at the Enteric Virus Unit.

Analysis conducted in order to match outbreaks recorded in the hospital outbreak database to date (n=1832) and those received at EVU for strains surveillance using the ILOG number as the outbreak unique identifier showed the following:

- A total of 80% of outbreaks reported through the hospital surveillance system lacked and ILOG number (therefore these samples could not be matched).
- Of the 20% of outbreaks for which an ILOG number was recorded, 16% could not be matched because either samples from the outbreaks were not received in EVU, or the samples were sent to EVU without the corresponding ILOG number.
- There were 76 outbreak records (4%) with an ILOG number that could be matched to a sample received in EVU with a corresponding ILOG number.

Figure 6: Proportion of samples in outbreak database matched to samples received in EVU. Samples were matched by taking the ILOG number as the identifier between the hospital outbreak database and the database of samples received in EVU.

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**BIBLIOGRAPHY**


