Norovirus Tracing in Environmental and Outbreak Settings – Experiences of waterborne, foodborne and nosocomial transmission

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av
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I. Nenonen NP, Hannoun C, Horal P, Hernroth B, Bergström T.
Tracing of norovirus outbreak strains in mussels collected near sewage effluents.
Applied Environmental Microbiology 2008; 74(8): 2544 – 2549.

II. Nenonen NP, Hannoun C, Olsson MB, Bergström T.
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III. Nenonen NP, Hannoun C, Larsson CU, Bergström T.
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IV. Nenonen NP, Hannoun C, Svensson L, Torén K, Andersson LM, Westin J, Bergström T.
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Journal of Clinical Microbiology 2014; 52(7): 2352 – 2358.

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Noroviruses (NoV), a major cause of acute gastroenteritis in hospital settings, also occur as sporadic infections or periodic non-seasonal community outbreaks. Human NoV replicates to high concentration in the intestinal tract, is readily transmitted by the faecal-oral route, hand-to-hand contact, contaminated food and water, and by aerosols. Large numbers of NoV are discharged into wastewaters and, despite sewage treatment, can cause problems when recycled river waters are used as source of drinking water. Two major groups of NoV are associated with human infections, genogroups (G) GI and GII. Epidemiological studies indicate association of GI with non-seasonal food- and waterborne infections, and GII with person-to-person transmission, particularly nosocomial spread of NoV.

As NoV detection in filter-feeding bivalves may have a sentinel role in tracing NoV in environmental waters, molecular tools were used to detect and characterize NoV in mussels from Fotö near the plume of sewage effluents from Gothenburg wastewater treatment plant. Sequence analyses of NoV RNA from Fotö mussels revealed GI.1 strains with high similarity (99%, 3.1kb) to strains detected in patients infected in non-seasonal, waterborne outbreaks linked to bathing in Lake Delsjö. Comparative sequence analysis of NoV strains from mussels and patients indicated that human NoV outbreak strains circulate in wastewaters, and can be traced in bivalves.

Molecular methods were used to characterize NoV detected in oysters implicated in a gastroenteric outbreak where only those guests who ate oysters were affected. Mixed human NoV GI and GII strains were found in the oysters, evidence of faecal contamination of the bivalves, held for several weeks in Strömstad harbour waters. NoV GI.1 strains from the oysters showed high similarity (≥ 99%, 285 nt) to the GI.1 detected in faeces obtained from one of the oyster-eating patients. Phylogenetic analyses of GI.1 strains from patient and oysters indicated the contaminated bivalves as point source of infection.

The similarity (99%, 3.1kb) of NoV GI.1 detected in Fotö mussels, patient samples from Delsjö waterborne outbreak, and the Strömstad oyster outbreak, was remarkable. High similarity held also when strains were compared with GenBank references; 96% with L23828, from an oyster outbreak, Japan 1989; 87% with the original Norwalk strain M87661, 1968, point source well water. These findings indicate genomic stability of NoV GI.1 strains over a period greater than 20 years, and dispersal of GI.1 in environmental waters.

Association of NoV GI strains with outbreaks related to sewage-contaminated water was emphasized in the molecular epidemiology of a large, non-seasonal waterborne outbreak affecting Lilla Edet, situated on the River Göta. Molecular studies revealed marked genomic diversity of NoV GI strains in patient samples. Cloning was used to confirm mixed GI infections including a new genotype, proposed NoV GI.9. Upstream sewage contamination of recycled river water and disinfection problems at the municipal drinking water treatment plant precipitated the outbreak.

In contrast study of NoV infections in hospital settings showed predominance of GII.4 strains in symptomatic patients and their environment. High similarity (≥99.5%, 1040 nt) was found between GII.4 variant strains from patients, and strains from dust, air, and surfaces in the patient's room. GII.4 strains detected in symptomatic patients in 8 wards during the 5-month study clustered on 11 sub-branches of the phylogenetic tree. One of the wards, a control, was not affected by nosocomial spread of NoV GII.4. High similarity of GII.4 strains from patients and their hospital room environment, in a given ward at a given time, confirmed nosocomial transmission and indicated the need for interventional cleaning studies.

To summarize, NoV tracing provided strong evidence of bioaccumulation of outbreak-related NoV strains in mussels growing near sewage effluents. High similarity of NoV strains from oysters implicated in a NoV outbreak and from an infected patient, indicated transmission of NoV from oysters to humans, confirming high stability of GI.1 strains in oysters, water and mussels. Cloning confirmed mixed NoV GI infections in patients from a waterborne outbreak, strengthening indices of an outbreak caused by sewage-contaminated drinking water. High similarity of NoV GII.4 strains detected in patients and their hospital room environment, confirmed local nosocomial transmission.

Keywords: Norovirus, tracing, environment, outbreak, waterborne, foodborne, nosocomial, mussels, oysters, GI.1, GII.4, dust, air, surfaces, molecular epidemiology

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