HUMAN NOROVIRUS

THE ORGANISM/TOXIN

Human noroviruses (NoV) are now the most common cause of outbreaks of epidemic non-bacterial gastroenteritis world-wide. Previously known as Norwalk-like viruses (NLVs) and small round structured viruses (SRSVs), these viruses belong to the *Caliciviridae* family and are 26-35 nm non-enveloped single stranded positive-strand RNA viruses. Noroviruses are divided into 5 genogroups, GI-V, of which GI, GII and GIV are known to infect humans. Over 25 different human genotypes are now recognised (Green, 2007). Since 2002, genotype GII.4 strains have been the most common cause of outbreaks. Recombinant NoV strains have also been identified (Koopmans, 2008; Greening and Wolf, 2010). Norwalk virus, GI.1, is the prototype strain. Norovirus identification has been difficult prior to development of molecular methods because human noroviruses cannot be cultured, and their wide genetic diversity limits the use of traditional immunology and serotyping assays.

GROWTH AND CONTROL

Growth

Human norovirus has not been grown *in vitro,* although infection of an organoid cell culture was described in 2007 but has not been reproduced since. There is still no suitable animal model. The murine norovirus strain is readily culturable. NoV detection is carried out using conventional or real time RT-PCR, with further characterisation or genotyping by DNA sequencing.

Survival

NoV survival data is mainly based on dose-response research studies carried out in the 1970's. These studies indicated that the virus was stable and resistant to heat, acid and solvents but no quantitative data is available. The virus retained infectivity after exposure to treatment with 20% ether at 4°C for 18h (Green 2007). Recent human dose-response studies show that the original Norwalk virus used in 1970's experiments was still infectious 25 years later (Teunis *et al.,* 2008).

Inactivation

Limited information is available (Koopmans and Duizer 2004). Knowledge of human NoV survival is based mainly on studies using environmentally challenged culturable surrogates (poliovirus, murine norovirus, hepatitis A virus and feline calicivirus), which have provided some inactivation data following treatment with disinfectants, heat, ultrahigh pressure and high and low pH (Duizer *et al.,* 2004; Carter, 2005; Koopmans and Duizer,2004; Hewitt and Greening, 2004). It is not valid to attribute these findings directly to human norovirus behaviour. Recent novel molecular approaches have provided some evidence of inactivation in response to temperature, relative humidity, heat and exposure to UV light (Lamhoujeb *et al.,* 2008; Wolf *et al.,* 2009a).

Temperature

The virus retained infectivity after incubation at 60°C for 30 min (Green, 2007). Pasteurisation is not sufficient to eliminate viruses. Resistance is reported to be greater in foods and shellfish. Steaming of bivalve shellfish is unlikely to inactivate NoV (Hewitt & Greening, 2006).
Under refrigeration and freezing conditions the virus remains intact and viable for several years.

pH

Resists gastric acids at pH 3-4. The virus retained infectivity after exposure to pH 2.7 for 3 hr at room temperature. Believed to be sensitive to pH >9.0 but unproven.

Water activity

Based on data for other enteric viruses and virus indicators, it is likely that NoV persist in waters for extended periods (possibly weeks/months) (Carter, 2005; Rzezutka and Cook 2004). NoV have caused many waterborne outbreaks and are often detected in environmental waters.

Drying

Resistant. Infectious NoV were detected on environmental surfaces, including carpets, for up to 12 days after NoV outbreaks (Carter, 2005; Greening and Wolf, 2010).

Preservatives
Unknown but likely to be resistant.

Sanitisers/Disinfectants
Dose-response studies showed that NoV was resistant to inactivation following treatment with free residual chlorine of 0.5 to 1.0 mg/mL. This level of free chlorine is consistent with that generally present in a drinking water supply. Did not cause infection after 30 min exposure to 10mg/L chlorine. (Green, 2007) Recent research suggests NoV may not be more resistant to chlorine than other enteric viruses (Carter, 2005).

Radiation
Evidence of norovirus inactivation following UV exposure (Wolf *et al.,* 2009a).

CLINICAL PICTURE

Incubation: 10-50 hours (mean 24h) following ingestion of the virus.
Symptoms: Vomiting, often projectile, is generally the predominant symptom and is present in > 50% of cases. Stomach cramps, watery non-bloody diarrhoea, abdominal pain, low-grade fever and headache are other common symptoms. The duration of illness is usually between 24-60 hr. Diagnostic criteria' are often used in the absence of virological confirmation (Green, 2007). Excretion of the virus in stools occurs from onset for up to 4 weeks following infection, with peak excretion rate at 4 days. Noroviruses are frequently discharged in vomit. The disease is generally mild and self-limiting. Hospitalisation is not generally required, but has been reported in some outbreaks. Attack rates are high, generally around 40-60% and sometimes as high as 80%.
Condition: Gastroenteritis. Norovirus colonise the proximal region of the small intestine and cause development of mucosal lesions with broadening and shortening of the microvilli. Short-term maladsorption of fats and some sugars has been reported. Abnormal gastric motor function is believed to be the cause of associated nausea and vomiting. The exact mechanism of pathogenesis remains unclear. Susceptibility or resistance to certain strains of HuNoV appears to be associated with human histo-blood group antigens (HBGA) Immunity is generally short lived, and is not sufficiently cross-reactive to protect against different norovirus strains.
Dose: Infective dose is estimated at 1-10 particles. Consumption of 1 virus particle may cause infection in 50% of occasions in susceptible people (Teunis et al., 2008). Viral levels shed in faeces can be >107/g.
At Risk Groups: Affects all age groups, but the elderly and the immuno-compromised are particularly susceptible.
Long Term Effects: There is no evidence of any long-term sequelae following norovirus infection. Fatalities, which mostly occur in elderly patients, are rare. US data assumes a case-fatality rate of around 11% of 2800 fatal cases of viral gastroenteritis per year.
Treatment: Usually none, but fluids may be given to reduce the risk of dehydration.

SOURCES

Human: The only known direct source for human NoV is human faeces. Other indirect sources are shellfish, contaminated foods, water, fomites and the environment.
Animal: Bovine, ovine and porcine noroviruses have been identified in New Zealand (Wolf *et al.,* 2009b) but there are no reports of cross-species transmission to humans as yet. Overseas, human GII NoV sequences have been identified in swine. Other caliciviruses have been found in various animal species.
Food: Contaminated bivalve shellfish, fresh produce (eg. berries, herbs, lettuce, salads), water, ice and manually prepared ready-to-eat foods (including bakery items). Poor hygiene practices by food harvesters, processors and food handlers are a significant source.
Environment: Faeces from infected humans may contaminate soil or water. Faecal pollution from sewage discharges, septic tank leachates and boat discharges has caused contamination of shellfish beds, recreational water, irrigation water and drinking water. NoV are believed to survive for long periods in the environment and have been detected in shellfish 8-10 weeks after contamination (Greening & Wolf, 2010).
Transmission Routes: The faecal/oral route is the established route of transmission. Infection occurs following ingestion of faecally-contaminated food and water. Another important route is person-to-person spread via aerosolised vomit following projectile vomiting. Direct transmission via contaminated surfaces, especially carpets, is also now considered a significant route (Greening & Wolf, 2010); These routes contribute to the explosive outbreaks that cannot be attributed to faecal/oral spread alone. They generally occur in semi-closed communities such as rest homes, cruise ships and camps where there is close quarter living and often there may be reduced hygiene levels. There is evidence that asymptomatic foodhandlers can also cause infection and may excrete high numbers of NoV in their faeces (Todd *et al.,* 2008).

OUTBREAKS AND INCIDENTS

In the United States, NoV infection is now classed as the major cause of foodborne disease, responsible for at least 9 million cases per year (Mead *et al.,* 1999). Because of the relatively high attack rate, large numbers of people are often infected and most cases of disease are outbreak-related. In New Zealand outbreaks are most commonly attributed to either foodborne or person-to-person contamination. Most foodborne outbreaks are ascribed to cross contamination via a foodhandler or inadequate cooking of previously contaminated foods. However, any food that becomes contaminated can act as a vehicle. Uncooked or lightly cooked bivalve shellfish such as oysters and mussels present a risk to health if grown in faecally contaminated waters.

NZ Incidence: NoV have been a leading cause of gastroenteritis outbreaks in New Zealand for many years. The overall attack rate in New Zealand outbreaks is estimated at 40-60%, but can be higher in institutional outbreaks and other settings as many cases are unreported. A recent study estimated 403,000 NoV infections occur annually in NZ (Cressey & Lake, 2008). Outbreaks occur throughout the year in New Zealand, which contrasts with that observed overseas, including Australia, where a seasonal peak occurs in winter. Surveillance data recorded in the database EpiSurv (http://www.surv.esr.cri.nz/surveillance/annual_outbreak.php) and reported in the ESR Annual Outbreak Summaries show there were 809 reported outbreaks of norovirus infection, including a total of 18,508 cases from 2001-2007. Of the total outbreaks, 19.9% (161/809) were associated with environmental sources, 17.6% (142/809) were associated with foodborne infection and 61.0% (494/809) were associated with person-to-person transmission. For several outbreaks, both person-to-person and foodborne transmission were recorded. There are no data on the incidence of sporadic NoV infection in the New Zealand.

Significant New Zealand Outbreaks

Shellfish: There have been a number of norovirus outbreaks linked to consumption of both commercially grown New Zealand shellfish and imported shellfish (Simmons *et al.,* 2001; Greening *et al.,* 2001; Simmons *et al.,* 2007). In 1999, contaminated commercially–grown NZ oysters were associated with 10 gastroenteritis outbreaks comprising 86 cases. The same strain of NoV was found in both oysters and faecal specimens of cases (Simmons *et al.*, 2001). In 2006, a large quantity of frozen imported South Korean oysters was served at an international Rugby match. Although labelled 'cook before consumption', the oysters were served raw and several hundred people became ill. NoV were identified in both the faecal specimens from cases and oysters (Simmons *et al.*, 2007). Several outbreaks linked to commercially grown NZ oysters from the same lease occurred in 2004 and 2008. NoV was identified in cases and oysters from both outbreaks.

Water: In 2001 and 2006, large NoV outbreaks were linked to consumption of sewage-contaminated drinking water at 2 NZ skifields (Greening *et al.,* 2001; Hewitt *et al.,* 2007). NoV GI.5 strains were identified in 11/31 cases and in both the source water and tap water in the 2006 outbreak (Hewitt *et al.,* 2007).

Foodhandling: There have been a number of NZ norovirus outbreaks implicating foodhandlers. In 1999, 62 people were infected with NoV at a buffet lunch in a hotel. Five foodhandlers had symptoms of norovirus infection when food was prepared before the event. In another restaurant outbreak, patrons from 4 different groups were ill and all had the same NoV strain as the implicated foodhandler (McLean *et al.* 2001).

Person-to-person: Person-to-person transmission is generally the predominant mode of spread in NZ. The majority of norovirus outbreaks occur in closed setting outbreaks, especially in rest homes, hospitals, child-care facilities and school camps.

Overseas outbreaks

There are many reports of foodborne norovirus outbreaks from overseas, where transmission has been associated with a wide range of foods, including bakery goods (bread rolls, wedding cake, icing), meat sandwiches, salads, shellfish, berries and contaminated water (Koopmans, 2008;Greening & Wolf, 2010).

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