QUANTITATIVE MICROBIAL RISK ASSESSMENT – RECENT ADVANCES IN NEW ZEALAND AND THEIR APPLICATION TO MOA POINT WWTP BYPASS DISCHARGES

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ABSTRACT

Quantitative Microbial Risk Analysis is increasingly being used for the assessment of Public health risk in the New Zealand water sector. It may now be considered a 'Best Practice' when assessing public health risk associated with wastewater discharges. Traditionally, the health risks associated with such discharges are assessed against faecal indicator bacteria concentrations (FIBs). These have been established from epidemiological studies that may not adequately mimic conditions, particularly near outfalls. QMRA, together with advancing enumerative laboratory techniques has allowed the outfall-associated public health risk assessment to focus directly on pathogens. Another significant benefit of QMRA methodology is that it allows the particular effect of an activity to be isolated from the gross effect of all similar activities in the same geographical space (e.g., stormwater discharges).

This paper, based around a recent QMRA of bypass flows from the Moa Point (Wellington City) Wastewater Treatment Plant (WWTP), discusses recent advances in the use of QMRA in New Zealand. In particular, a) the adoption of human Norovirus as a key risk organism b) derivation of a non-point source model for the risk of respiratory illness and c) the move from 'Risk of Infection to 'Risk of Illness' assessments.

We also discuss issues that have arisen that will require further research as this powerful risk assessment methodology gains further acceptance and, as is already the case in some situations, 'required' status.

KEYWORDS

Quantitative Microbial Risk Assessment, Norovirus, Adenovirus, Disinfection, Dose Response

1 INTRODUCTION

1.1 QUANTITATIVE MICROBIAL RISK ASSESSMENT

Quantitative Microbial Risk Assessment (QMRA) has increasingly come to be regarded as a component of 'Best Practice' in public health risk assessment associated with wastewater discharges. These are:

- A growing public demand to 'know' the health risks, from a particular discharge, as a result of pathogenic organisms that are thought to possibly pose greater risk than is reflected by the traditional faecal indicator bacteria (FIB).
- Where multiple sources of risk exist, the ability to isolate the likely effects of each such that appropriate corrective action/s can be prioritised. This is in contrast to field studies which can be lengthy, expensive and often inconclusive as to the effects of a particular discharge activity.
- Advancing laboratory methodologies for enumeration of viral and protozoan pathogens
- Clinical trials and outbreak studies have investigated a wider range of pathogens and a documented a range of human responses to various doses of those pathogens

QMRA is not intended to replace field analysis but to supplement it and improve the outcomes suggested by field work.

Because the use of QMRA is increasing in parallel with continuing advancements in the associated science, we are not only able to improve the accuracy of assessments with each successive use of the technique, but we also tend to find new limitations and or shortcomings for which improved science is required or for which modelling 'work-arounds' are required until the science is more fully developed. In this most recent Moa Point WWTP QMRA, the following such issues have arisen, all of which require further research and development:

- The RT-qPCR methodology developed recently in New Zealand, by ESR, for enumerating Noroviruses, tells us the number of individual replicates of the virus as opposed to the 'aggregated' enumeration used by Teunis his clinical trials and subsequent development of the dose response model currently used for assessing the 'risk of infection'.

- The 'apparent' lack of physical removal of Norovirus through filtering, adsorption or encapsulation in the suspended carrier and suspended growth reactors.

- The inability to culture human Norovirus in the laboratory meant that the efficacy of the UV disinfection system in inactivating Norovirus, could not be measured directly, as opposed to Adenovirus (used for respiratory illness risk assessment) which could be measured across the entire plant.

- The lack of a useable model for estimating the generation and transport of atmospheric viruses from the two dimensional sea surface, given a concentration of the virus in the marine water.

1.2 MOA POINT WASTEWATER TREATMENT PLANT BYPASSES

Moa Point WWTP treats municipal wastewater for the majority of Wellington City, serving some 195,500 people. The treatment plant has resource consent to discharge partially treated wastewater bypass flows from the Moa Point outfall if required. Condition 4 of the resource consent (WGN080003 [26181] issued in May 2009) requires that a 0.5 log reduction of enteric viruses is achieved at all times, once a UV disinfection facility has been commissioned. Condition 15 of that consent required an investigation into using ultra violet (UV) disinfection to obtain the 0.5 log reduction of enteric viruses. A pilot study (Veolia Water 2012) and investigation of disinfection performance, during bypass events, concluded that using UV disinfection, at the Moa Point WWTP site, is unlikely to achieve the 0.5 log reduction required 'at all times' during events in which a portion of the raw sewage bypasses the main treatment 'train' before blending with the treated effluent to discharge through the long sea outfall.

Alternative options were investigated by Veolia Water which concluded:

- To achieve the required reduction of enteric viruses, in the bypass flow that a larger [UV Disinfection] system would be required at an estimated cost of over $20M CAPEX and an estimated $1M OPEX.
- That the required reduction, between influent and effluent, is already achieved due to the 6:1 dilution effect of mixing the fully treated discharge with the partially treated discharge (Veolia Water 2012).

Capacity Infrastructure Services, on behalf of Wellington City Council was required to assess the effects of occasional sewage bypass events from the WWTP, under resource consent conditions that were placed on the WWTP to alleviate public concerns about the health risks associated with bypass discharges.

Bypass events have numbered 48 since January 2001. In that time, bypass conditions have existed for a total of 0.2% of the time. The prescribed methodology was a shellfish survey along the affected coastline. For a number of reasons (including lack of credible shellfish populations, timing between discharge and effect, safety and inability to isolate bypass effects from those of other activities (including compliant discharge)) this methodology would not have provided useful results.

An alternative methodology, using QMRA, was offered and adopted. This involved an influent / effluent virus monitoring programme, 2D dispersion / dilution modelling of the near field Cook Straight area and Monte-Carlo style risk modelling for beach goers at 6 sites considering swimming, surfing, shellfish consumption and non-water based activity. The assessments covered the risks of contracting both gastrointestinal and respiratory illness, specifically as a result of the bypass events contaminating the air, seawater and shellfish flesh.

The description of the Moa Point QMRA study presented in this paper can be referenced in full in Crawford and Bell (2013).

2 QUANTITATIVE MICROBIAL RISK ASSESSMENT

2.1 THE PRINCIPLE

QMRA works by statistically modelling known and estimated distributions along the source to receptor pathway that includes influent pathogens, dilutions, treatment plant performance, dispersion and inactivation in the receiving environment, ingestion by water users and the response of individuals to doses of differing sizes.

This technique recognises that pathogen loads from the population vary and that any particular process that acts upon the viruses can produce a range of kill or inactivation rates. This is as opposed to older styles of risk assessment which adopted representative single numbers for the pathogen numbers and the rate of kill, inactivation or dilution at each stage to derive a final 'risk number' then apply sensitivity analysis to pick up the boundaries of probability.

2.2 GENERAL METHODOLOGY

The generalized QMRA methodology is described in Haas et al. (1999) and, more specifically in papers such as McBride et al. (2005). The following provides a very brief summary of the methodology.

In this approach all available information is used to define the environmental proliferation of the pathogenic organisms, their fate through the treatment plant and receiving environments and dose-response relationships developed from clinical trials and from outbreak data, in five steps:

- *Hazard identification*: Which ones (a hazard is a quantity that has a potential to cause harm)?
- *Exposure assessment*: What exposure might a population have to a hazard (via water contact recreation or by consumption of raw shellfish harvested from water containing the hazard)?
- *Dose-response analysis*: What is the probability of infection (or illness) given ingestion or inhalation of one or more pathogen particles?
- *Risk characterization*: How much infection or illness would arise in a population exposed to a distribution of pathogens in the water or shellfish? What is the risk of illness faced by an exposed individual?
- *Risk communication*: Necessary for a clear understanding of the meaning of the risks that have been identified.

Figures 1 and 2 represent the source to receptor pathways that have were chosen for the Moa Point risk assessment and show the main points at which there is significant variation in the parameters that are to be modelled.

3 MOA POINT WWTP QMRA

3.1 OBJECTIVES

The objectives of the QMRA based investigation were to:

1. Investigate the public health risks at the locations of frequent human contact, right through to the individual's illness risk (IIR) under recognised ranges of human activity and using recognised dose response models.

2. Isolate the risks associated with pathogens discharged from the treatment plant from those originating from background sources such as stormwater run-off, animals and other wastewater discharges.

3. Investigate the risks under as wide a range of discharge scenarios as possible.

4. Investigate the risks under as wide a range of system input conditions as possible based on monitoring a range of actual inputs over time and the use of information from other locations regarding possible extreme events (for example the outbreak of a viral infection epidemic in the community coinciding with a treatment plant bypass event).

3.2 BYPASS CONTEXT

Average daily flow to the treatment plant is 72,000 m^3/d. The instantaneous capacity of the biological and disinfection components of the treatment plant is 3.0 m^3/s. The instantaneous flow capacity of the treatment plant feed pumps and screens is 4.5 m^3/s. Therefore, under extreme flow conditions, up to one third of the flow

arriving at the treatment plant can bypass the main treatment elements. Under normal circumstances, up to 3 m^3/s receives full treatment.

Bypasses are almost always due to the influent to the treatment plant exceeding the instantaneous flow capacity of the main treatment plant processes, which is 3,000 litres per second (or 3.0 m^3/s). Rarely, it could also be due to a malfunction of the inlet screening facility. The maximum pumping capacity to the treatment plant is 4.5 m^3/s. Thus, the maximum bypass flow rate is 1.5 m^3/s. As the bypass flow rate increases, the overall treatment plant performance (after blending of the flows) is likely to decrease.

Since 1 January 2001, 48 bypass events have been recorded. The bypasses have been for a total of 188 hours and 26 minutes. Thus, full treatment is provided for approximately 99.8% of the time. As such, the levels of public health risk identified in this investigation, as being a result of bypass events, are only present a small proportion of the time. Table 1 provides some statistics on the bypass events.

Table 1: Bypass Statistics Since 2001

	Bypass events per year	Duration of Bypass hh:mm:ss	Bypass Volume (m^3)	Per cent of daily flow (%)
Average	4	06:23:00	5,065	3.03
Minimum	1	00:00:30	3	0.003
95th %ile		15:45:00	29,973	11.5
Maximum	8	18:03:00	45,500	43.5

3.3 SPECIFIC METHODOLOGY

The following subsections provide an abridged version of the methodology discussion from the main QMRA report (Crawford & Bell 2013).

3.3.1 MODELLED PATHWAYS

The pathways modelled for infection by the 'Hazard' were:

a) Gastrointestinal (Gi) illness via ingestion of sea water while swimming at:

- Houghton Bay / Princess Bay (Site 1)
- Dorrie Leslie Park (Site 2)
- Lyall Bay opposite the surf club (Site 3)
- Breaker Bay to the north (Site 6)

b) Gastrointestinal illness via ingestion of sea water while Surfing. It was recognised that the risks in areas used by surfers may be different to those for swimmers so a surf zone location was adopted in Lyall Bay where surfers are more likely to congregate (Site 4).

c) Gastrointestinal illness via ingestion of viruses in raw shellfish meat. This part of the modelling is somewhat hypothetical as there are no known, well established, viable beds of filter feeding bivalve shellfish in the area. There may however be established colonies of Paua.

d) Respiratory infection via inhalation of viruses in the air mass driven off contaminated marine water.

Figure 1: Location of the outfall and diffuser in Lyall Bay and specified model output sites.

The diamond also shows location of the recording current meter (RCM) moored in 1989 for the original assessment of environmental effects investigations and used for verifying the hydrodynamic model. [Image: Google Earth and DigiGlobe]

Figures 2 and 3 below represent the source to receptor pathways that have were chosen for the Moa Point risk assessment and show the main points at which there is significant variation in the parameters that are to be modelled.

3.3.2 HAZARD IDENTIFICATION

Human Norovirus of Genotypes GI & GII were adopted as the risk indicators for gastrointestinal illness. Key stakeholders considered, and agreed with, contemporary thinking that Norovirus should be measured and modelled as the key risk organism used in the bathing water and shellfish study. That is, the use of Norovirus over bacterial indicators, protozoa and other viruses. This is due to the prevalence of Norovirus as a contributor to gastrointestinal illness in New Zealand and the very highly infective nature of the organism. Greening et al. (2010) reported 'human Noroviruses are the most common cause of outbreaks of epidemic non-bacterial gastroenteritis worldwide (Siebenga et al. 2009) and that frequent Norovirus contamination of New Zealand shellfish (both commercial and wild) occurs, with over 50 Norovirus outbreaks, linked to consumption of either New Zealand commercially grown oysters or imported oysters, having been reported between 1994 and 2010.

Further, international literature has recently identified Norovirus as the likely main aetiological agent for recreational/shellfish waters (Sinclair et al. 2009, Soller et al. 2010). Soller et al. considered a range of bacterial, protozoan and viral pathogens in their work and found Norovirus to provide, by far, the highest risks of both infection and illness amongst swimmers in contaminated water.

Similarly, human Adenoviruses (HAdV) of serotypes 2, 3 and 4 were adopted as the key risk indicators for respiratory illness in beach goers, including surfers. Viruses are thought to be one of the most common sources of respiratory illness. Mims et al. (2004) reported that up to 70% of sore throats are caused by viral infections. Adenoviruses and Enteroviruses are typically associated with AFRI[1] symptoms and commonly encountered. Both are regularly measured during Moa Point bypass events. Adenoviruses are more robust in the environment than many other viruses[2] and have been associated with disease outbreaks from recreational waters (Sinclair et al. 2009). Of the range of Adenoviruses, we need to look specifically at the Human Adenoviruses (HAdV). The HAdV viruses present risks of both gastrointestinal and respiratory illness. HAdV serotype 4 has been linked to "persistent epidemics of acute respiratory disease in the US" and "Types 2 and 3 are generally associated with pneumonia and childhood respiratory diseases" (Fong et al. 2010). Thus we have concentrated on HAdV serotypes 2,3 and 4 as the respiratory serotypes and with an aggregate of up to 10% of the HAdV present, but most likely 3% (Fong et al. 2010, Kundu et al. 2013) (as we don't expect them all to be present at their maximum proportions at the same time). The pathway for Adenovirus infection and illness was inhalation of aerosols, generated from seawater potentially containing the virus, due to the action of wind over the water (on-shore breeze) and or the action of breaking waves.

[1] Acute Febrile Respiratory Illness. Generic term for illnesses where a person develops a fever of 38°C or more and a cough or shortness of breath.
[2] See IUVA (2006) for published dose rates for inactivation.

Figure 2: Modelled Virus Transport and Inactivation Pathway

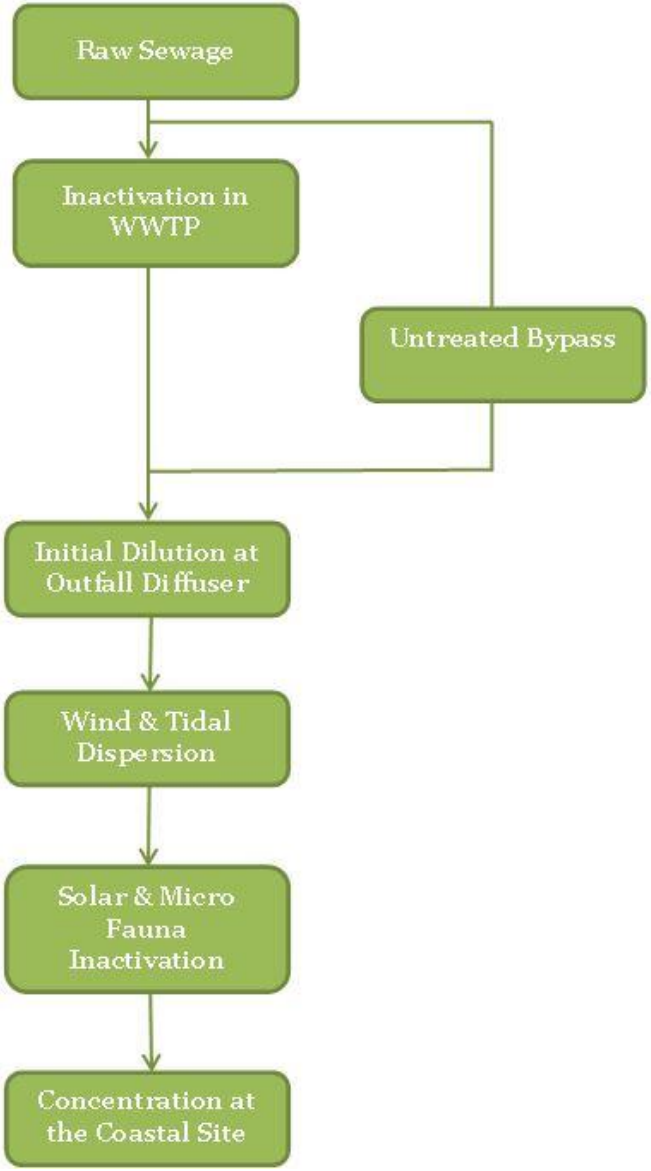
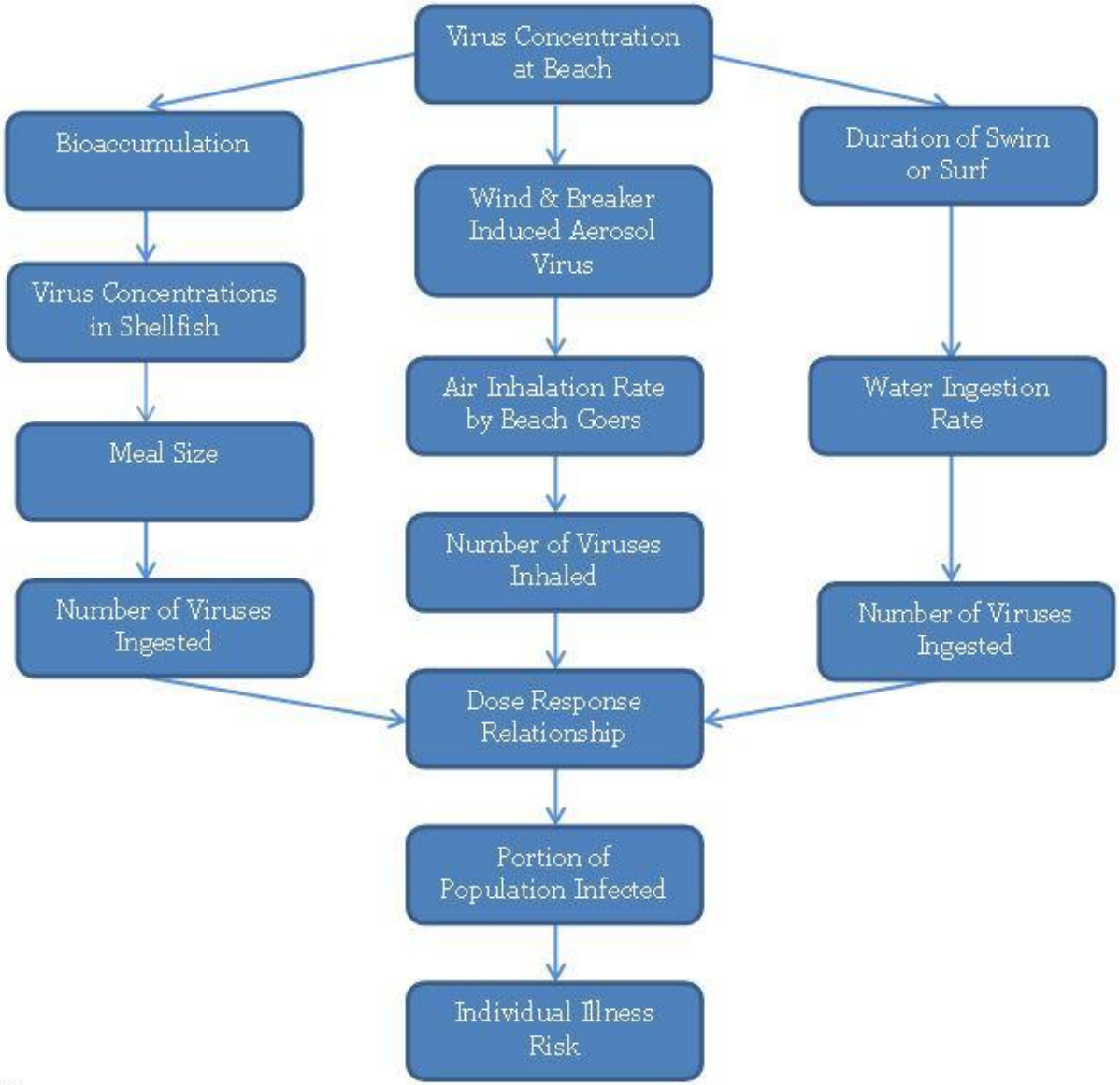


Figure 3: Modelled Infection & Illness Pathways



3.3.3 DATA COLLECTION

INFLUENT

During the study period, influent and effluent NoV GI & GII were measured on seven normal, 'Business as Usual' days and at intervals during a significant wet weather bypass event on 6 May 2013. There was no significant difference noted between the influent NoV concentrations on dry days compared to the 'very' wet weather day. However, this is not terribly surprising, because of the vast numbers of viruses present, significant differences in concentration are measured on a $\log_{10}$ scale, i.e. in factors of 10. Whereas an extreme wet weather event in the sewerage resulted in only a 3 fold increase in inflow volume. For example, whereas average day flow is 72,000 m^3/d, the 6 May 2013 event resulted in a flow of approximately 210,000 m^3. By contrast the range of influent NoV GI & GII during dry weather monitoring was from 2.1×10^4 to 3.5×10^5, whereas the average of 6 influent samples during the bypass event of 6 May rendered an average of 2.6×10^4 and a range from 1.6×10^4 to 3.8×10^4. Therefore, for the purpose of all QMRA modelling (except the two specific bypass events), the same distribution influent concentrations were used. The 8 data points from this study were supplemented with a further 6 Wellington influent / effluent sample pairs reported in Hewitt et al. (2011). Importantly, these included at least one 'non-detect' for each virus type, which meant that the lower bounds of each influent distribution could be reliably set at 'zero'.

To define the upper bounds of the influent virus concentration we drew on international data because our limited sampling programme could not hope to capture this. Lodder et al. (1999) monitored Norwalk like caliciviruses (NLV) in WWTP influent during three 'outbreak[3]' events in the Netherlands during 1997 and 1998. They measured raw sewage NLV concentrations up to 1×10^7 RNA containing particles (virions) per litre. Thus, we have adopted 1×10^7 as the upper bound or 100^{th} percentile for the distribution. Because of the limited number of samples, rather than fitting a continuous distribution to the influent NoV GI & GII results, we fitted a 'hockey stick' type distribution as described in McBride (2005a).

EFFLUENT

The **effluent Norovirus** (NoV) numbers were very similar to influent, both under dry and wet weather conditions. The RT-PCR technique available for NoV simply enumerates the single strand RNA particles. It cannot differentiate viable from non-viable. This indicated that the Moa Point WWTP is not particularly effective at physically removing NoV particles, which is somewhat surprising given the combination of settlement and floc-forming processes.

From the work of Teunis et al. (2008), the assumption is that the current dose response model is based on viable replicates of the virus found in raw, untreated sewage (see discussion in Crawford et al. 2013). However, even if little or no virus is physically removed by the treatment plant, the UV disinfection system will provide further inactivation relative to the UV dose applied.

Veolia Water, operators of the WWTP, measured UV applied doses of between 28 and 34 mWs/cm^2 during dry weather and between 22 and 29 mWs/cm^2 during the 6 May wet weather event. From the International Ultraviolet Association published Biodosimetric data[4] (IUVA 2006, Table 4) data for various caliciviruses[5], we can therefore safely adopt a most likely inactivation of NoV, for water that passes through the UV systems (as opposed to bypasses), of 3 $\log_{10}$ and a probable maximum of 4$\log_{10}$.

Two methodologies were available to assess the amount of HAdV inactivation to be modelled.

First, HAdV can be cultured in the laboratory (Noroviruses can't be cultured) so both the viable influent and viable effluent virus particles can be calculated (in this case, on average, only 1 in every 1,260 influent HAdV particles was also viable). The mean and median measured HAdV inactivation during the 2013 investigation was 0.7$\log_{10}$.

Second the IUVA Biodosimetric data (IUVA 2006) indicate the Adenoviruses are substantially more robust than other viruses, requiring approximately 40 mWs/cm^2 of UV_{254} dose per $\log_{10}$ inactivation. With a calculated average UV dose rate of 29 mWs/cm^2, the Moa Point WWTP performance corresponds to approximately 0.7

[3] Outbreaks of viral gastroenteritis have been epidemiologically linked with NLVs. The outbreaks occurred in nursing homes in the cities of Reeuwijk, Apeldoorn, and Enkhuizen.

[4] Note that the IUVA data is based on 'cultured' assays and this may be one reason why Noroviruses are not specifically included.

[5] Noroviruses are a form of calicivirus, small round viruses.

log_{10} inactivation. This is consistent with the 2013 measured inactivation and was adopted as the most likely inactivation with upper and lower limits of $1log_{10}$ and 0.0 respectively.

3.3.4 HYDRODYNAMIC DILUTION / DISPERSION MODELLING

Dispersion of by-pass discharges through the Lyall Bay long outfall were undertaken using a combination of 3 models or algorithms[6]:

A. two near-field models (CORMIX and DIFFUSER) to predict initial dilution in the vicinity of the outfall diffuser.

B. far-field hydrodynamic/dispersion model (Delft2d) for simulating physical dispersion achieved at each of the 7 coastal sites. A pre-existing, calibrated, 2d hydrodynamic model set up was able to be employed, minimal updating saving a considerable amount of time and money.

C. an algorithm to calculate virus inactivation in seawater based on using hourly solar radiation measurements from the Kelburn weather station.

Three different modes were utilised to generate concentration-reduction factors from the models for the QMRA procedure.

1. A 1-year long simulation to provide a full range of tide, wind and seasonal conditions (including solar radiation) so a comprehensive probability distribution of concentration-reduction factors could be determined, assuming a higher-discharge, by-pass event could occur at any time within the 12-month period 1 September 2011 to 31 August 2012. This 'synthesised event' drew on all previous discharge event data and assumed a continuous discharge condition over the entire year.
2. Based on the 1-year long simulation over the same period (1 September 2011 to 31 August 2012), to provide a probability distribution of concentration-reduction factors, at the specified coastal sites, for the WWTP discharge operating continuously on an Average Daily Flow (ADF) basis of 0.83 m^3/s.
3. Modelling in more detail the plume movement and concentrations for two particular by-pass events following heavy rain (3 March2012 and the largest event to date on 6 May2013) to assess the timing, extent and persistence of elevated concentrations at specified sites, and as a cross check that the generalised 1-year simulation is producing plausible, realistic results.

Some key findings from the 1-year generalised bypass simulation:

- Initial dilution within the vicinity of the outfall diffuser contributes the most towards reducing concentrations at each of the coastal sites, followed by subsequent dilution as the plume is transported and dispersed. The smallest reduction is from inactivation of viruses, particularly at night and during winter months when solar radiation is lowest.

- The median initial dilution is around 300-fold dilution or a concentration reduction factor of 0.0033 for the Average Daily Flow (0.83 m^3/s), but dilution decreases substantially as the effluent discharge increases, as plume mixing is much more efficient for lower discharges into high current speeds.

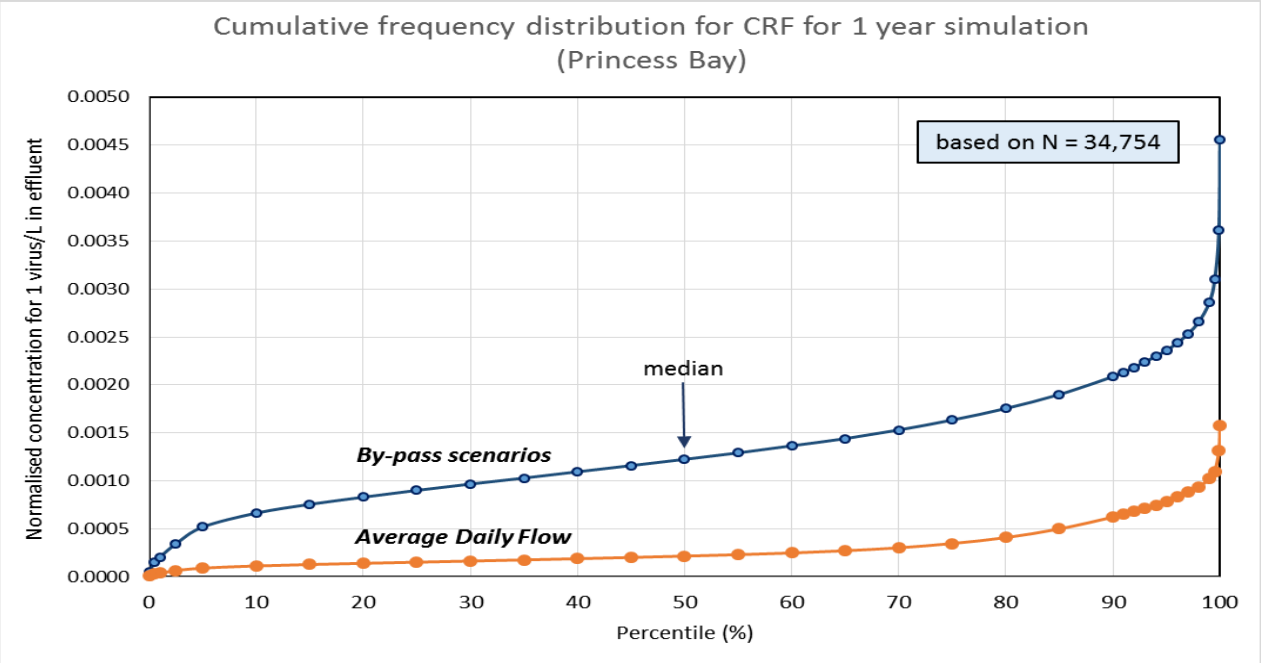
- Mostly, the subsequent-dilution factor ranges from 0.15 (or ~7-fold dilution) and occasionally higher, for sites in more direct line of travel for the diluting plume, down to 0.05 or less (or >20-fold dilution) for the furthest sites like Princess Bay and Breaker Bay. The median factor for Lyall Bay sites was just over 0.1, equivalent to a subsequent dilution of 10-fold.

[6] Three different dilution/decay models were used as described in Appendix D of Crawford & Bell (2013) validation was as follows:

- CORMIX (internationally accepted initial dilution that has been verified for a number of outfall sites around the world – so is not normally verified locally in applications)
- Delft2D tracer dispersion module (which only simulates dispersion of conservative or non-decaying substances). We used the dispersion coefficients used in the 1989 dispersion model investigations in Lyall Bay that were based on dye-tracking surveys.
- Microbial decay/inactivation – decay parameters based on microbial survival experiments as outlined in Appendix A of Appendix D.

- Median reduction factors for virus inactivation over the entire year ranged in a narrow band from 0.88 (Breaker Bay) to 0.92 (Hue te Taka and Princess Bay), which is equivalent to a median "dilution" of only 1.09 to 1.14-fold. In peak summer mid-day periods, the reduction factors for virus inactivation reduced to a minimum around 0.66 to 0.78.

Figure 4: Cumulative Distribution of the normalised concentration-reduction factor (CRF) for Princess Bay for a 1-year simulation, comparing by-pass scenarios with the constant ADF simulation



3.3.5 DETERMINATION OF ILLNESS RISK

With the temporal concentrations of the subject viruses in the areas of interest determined, the individual's illness risk (IRR) over a bathing season, or a year of shellfish gathering, was then modelled using a Monte Carlo style sampling routine (using the @RISK plug-in to Excel). The models sample environmental concentration, length or extent of exposure, rates of ingestion, dose response and probability of illness to derive distributions of potential illness risk outcomes.

For each exposure occasion, 100 separate individuals were exposed to allow the range of potential dose responses (of the individual to a particular dose) to be covered. For each exposure pathway, 10,000 exposure occasions were modelled (sampled) from the viral concentration data. This was to ensure that the full range of potential exposure was experienced by each individual with varying susceptibility. That is, through the full range of potential marine concentrations and plausible ingestion / inhalation quantities.

A summary of the dose response model parameters employed is provided in Appendix A.

3.3.6 SUMMARY OF MOA POINT BYPASS RISK RESULTS

The following points provide a very brief summary of the results of the monitoring programme and subsequent health risk modelling:

- Cumulatively for all beaches, the IIR (Gastrointestinal) from the combination of NoV GI & GII from Moa Point WWTP is less than 1%. The risk of illness is typically absent for around 80% of the duration of the event and its time to dissipate. The seasonal averaged risk of illness from NoV GI&GII, resulting from bypass events, is less than the 'tolerable risks' for 'A' Grade Microbiological Assessment category in NZ MoH/MfE (2003), and it is lower than the 3.6% and 3.3% recommendations of the USEPA (2012).

- For all beaches, apart from the Lyall Bay surf zone, the IIR (respiratory[7]) from human Adenoviruses is less than 0.3%. The risk is higher for surfers because this is not measured on a beach and they are

[7] For respiratory infection, the pathogens are breathed in with air.

exposed to aerosols generated by wind from any direction, not just the on-shore winds experienced by people sitting, walking or exercising at or in the water's edge.

- If there was a viable population of filter feeding shellfish on Hue te Taka Peninsula, they would not be regarded as safe to eat. During every day operation of the treatment plant, the IIR would be approximately 5%. This is elevated to 8% during and following a bypass event. The risk levels for the west side would be higher than for the modelled east side of the peninsula.

- The generalised bypass event model produced results worse than those for specific events, indicating the principal conclusions from the modelling are likely to be conservative, for the scenarios considered or, if the model is extended for use under different sites or scenarios.

4 RECENT ADVANCES

4.1 NOROVIRUS AS HIGHEST RISK VIRAL PATHOGEN

Historically, viruses such as Adenovirus, Rotavirus, Enterovirus (McBride et al. 2005, Crawford 2009) have been used in these QMRA assessments. These are of moderate to high infectivity, relatively common and comparatively easily enumerated. It was known that Noroviruses are very infectious. However, early quantitation methodology and a dose response model (Teunis et al. 2008) only became available in 2008 (Teunis et al. 2008). Both quantitation and dose response understanding for Norovirus have continued to evolve since then.

Now that it can be readily quantitated[8] and dose responses calculated in clinical trials (Teunis et al. 2008) and in outbreak studies (Thebault et al. 2013), Norovirus has been adopted due to its prevalence as a contributor to gastrointestinal illness in New Zealand and the very highly infective nature of the organism.[9] Greening et al. (2010) reported 'human Noroviruses are the most common cause of outbreaks of epidemic non-bacterial gastroenteritis worldwide' and that frequent Norovirus contamination of New Zealand shellfish (both commercial and wild) occurs, with over 50 Norovirus outbreaks, linked to consumption of either New Zealand commercially grown oysters or imported oysters, having been reported between 1994 and 2010.

Further international literature has recently identified Norovirus as the likely main aetiological agent for recreational/shellfish waters (Sinclair et al. 2009 & Soller et al. 2010). Soller et al. considered a range of bacterial, protozoan and viral pathogens in their work and found Norovirus to provide, by far, the highest risks of both infection and illness amongst swimmers in contaminated water.

4.2 RISK OF ILLNESS vs RISK OF INFECTION

Thebault et al. (2013) have published a Norovirus illness outbreak study in southern France (for consumers of raw shellfish) which has confirmed that the probability of this illness given that infection has occurred is rather high. Conversely, the earlier clinical trial study of Teunis et al. (2008) indicated that this probability is low (thousands of virions being needed to cause 50% of exposed susceptibles to become ill). The latter result has been treated with some skepticism, as it appears to be at odds with observed food-related Norovirus illness outbreaks. Current practice (e.g., Soller et al 2010) is to treat this probability as a dose-independent number, and we have followed this practice, as explained in the Appendix.

4.3 AIRBORNE PATHOGEN GENERATION FROM A 2D WATER SURFACE

For the Moa Point QMRA study, no readily usable data or model was available to estimate or simulate the concentration of viruses in the general air mass (as opposed to individual aerosol particles) blown above the above and off the contaminated water mass. While there is a reasonable body of work considering the amplification on concentrations in the mass water body up to those in the individual aerosol droplet, this does not translate to concentration in the general air mass. Further, there was available, a one dimensional model for the generation of atmospheric pathogens as used by McBride & Stott (2011). However, this was only applicable to a point source of pathogens rather than a 2 dimensional surface.

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[8] ESR at Kenepuru has developed a reliable RT-qPCR technique which gives the non-aggregated enumeration.

[9] A recent study by Atmar et al. (2013) has challenged the infectivity status of Norovirus. However counter-arguments (McBride 2014, Messner 2014) have pointed in the opposite direction: Norovirus remains an extremely infectious pathogen to a susceptible proportion of a population.

We were eventually able to develop plausible and realistic relationships (between wind speed an resulting atmospheric concentration), using actual measured data for salt concentration vs wind speed (Blanchard 1982) and for 'red tide' algal blooms in the Gulf of Mexico where Yung et al. (2005) and Pierce et al. (2005) measured near surface air mass brevotoxin concentrations vs wind speed. Importantly, they measured the brevotoxin concentrations in the sea water and in the air mass. A full description of the respiratory model derivation is provided in appendix B of Crawford & Bell (2013).

5 NEED FOR RESEACH AND DEVELOPMENT

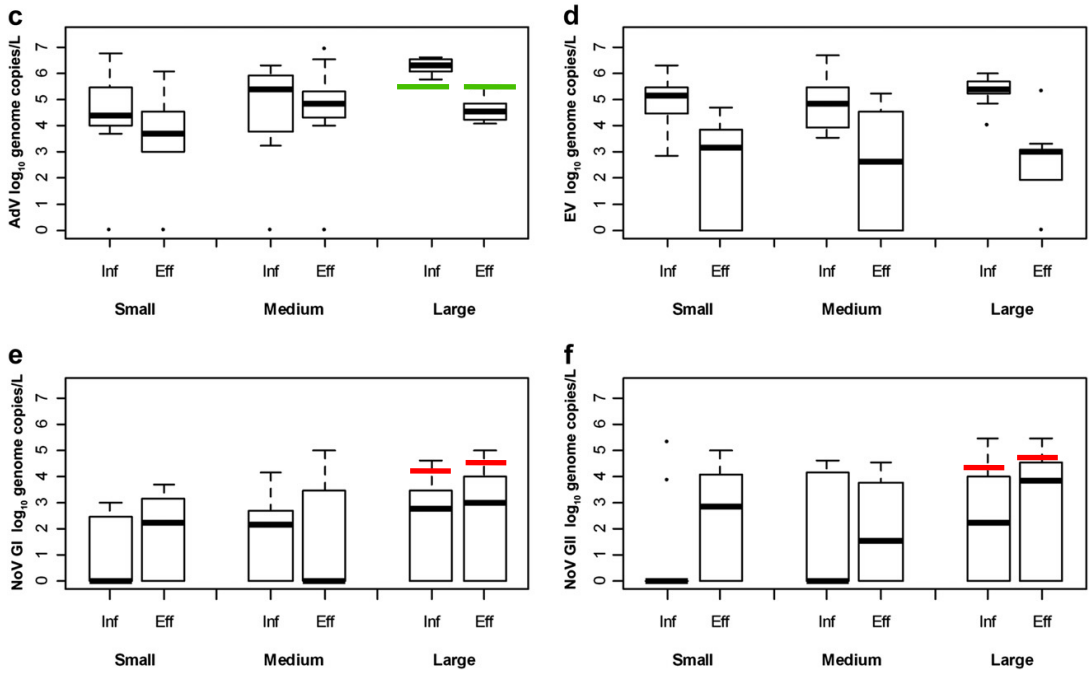
5.1 NOROVIRUS AGGREGATION ISSUE

Dose-response functions for viruses, as used in sewage-related QMRA studies, are based on clinical trial studies and account for the possible presence of aggregation of the viruses (Messner 2014, McBride 2014). That accounting *increases* the infection and illness probabilities. However, some treatment processes aim to aggregate virions,[10] and so the aggregation-free approach is always precautionary. That of course is an appropriate stance to take when considering public health risks. Nevertheless, a better understanding of the role of aggregation would be helpful in refining the QMRA approach.

5.2 WHY ISN'T NOROVIRUS PHYSICALLY REDUCING THROUGH TREATMENT PLANTS?

When Norovirus and Adenovirus quantitation by PCR methods (measures physical presence, not viability) were considered, we noted that even though Moa Point WWTP employs a suspended growth media phase and a suspended growth phase followed by secondary clarification, there appeared to be little, if any physical removal of Norovirus or Adenovirus particles from the sewage between plant entry and exist. This finding was not what we would have expected from such a treatment plant but (with regard to Norovirus) was consistent with the findings of Hewitt et al. (2011) across a range of New Zealand WWTPs. This is concerning for when considering Norovirus, due to its prevalence and high infectivity. While there was virtually no difference in the median Adenovirus quantitation by PCR across the plant, there was an appreciable difference in the average.

Figure 5: Reproduction of part Fig 1 from Hewitt et al. 2011 (Published by Water Research) (those results of PCR analysis) for ten NZ WWTPs. Thick black bars are medians. Note Adenovirus and Enterovirus particle numbers decrease whereas Norovirus GI & GII particle numbers increase through the treatment plant. Moa Point Norovirus PCR median results are shown in red, again increasing. Moa Point Adenovirus PCR median results are shown in green.



[10] For example, coagulation could promote aggregation; but filtration may have the opposite effect.

Because the main processes employed form sticky 'flocs' which tend to mop up other matter and, because the flocs are then separated from the effluent in a gravity settlement phase, we would, intuitively, expect that there would be consequent physical removal of Norovirus as is known to be the case for bacteria and other viruses.

This is an important public health issue for our treatment plants and warrants further research, particularly if the current trend toward consideration of viruses, rather the faecal indicator bacteria, is to continue.

5.3 AIRBORNE PATHOGEN GENERATION FROM A 2D WATER SURFACE

While we were able to develop plausible relationships for the sea to air transfer of pathogens with changing wind speed, the relationships were inferred and none of the reference research was directly made on any virus, let alone the high risk species we were considering. We consider that, if respiratory risk assessment is to become a common aspect of QMRA studies, direct research would be very valuable in deriving direct relationships for the viruses we wish to study. We consider that this research could be made as a combination of field studies under measured conditions and very controlled conditions in a wind tunnel facility using a water body spiked with target organisms at varying concentrations for various wind speeds.

5.4 BIOACCUMULATION OF VIRUSES IN BI-VALVE MOLLUSC TISSUE

Currently, when QMRA studies are assessing the risks due to ingestion of raw shellfish tissue, bi-valve molluscs are used as the vector because they are very common and accessible in New Zealand waters, are very frequently consumed raw and because they are known to 'bioaccumulate' pathogens. Currently, a relatively coarse bioaccumulation approach is employed. This is conservative, in that uptake is assumed to be instantaneous, but depuration is underestimated. The resulting risk profiles are plausible, in the sense that risks from raw shellfish consumption are always calculated to be rather higher than risks associated with swimming in or near to the shellfish-harvesting waters. Nevertheless, better shellfish kinetics models are needed, and are under development.

6 CONCLUSIONS

QMRA processes such as described here have a number of advantages over conventional public health risk assessment methodologies, in that they:

- focus on particular, high risk, target pathogens in the actual environment concerned, rather than relying on results from epidemiological studies in dissimilar environment conditions;
- allow the comparison of alternative wastewater treatment methodologies and discharge location and configuration options;
- provide for calculation of risks attributable to particular sources. Thus isolating the health risk of a particular activity from the risks resulting from other activities in the same geographical area that also contribute to the same hazard/s;
- allow risk assessments to be made over a wide range of environmental conditions without having to wait for (and have sampling teams on standby for) ideal conditions representing each condition that needs to be assessed;
- quantify risk outcomes in terms that are easily understood by the public.

Nevertheless, sewage-related QMRA is, as yet, a young science topic, with a number of ongoing research needs such as:

- better characterization of the probability of illness, through further clinical trials, and refinement of the existing dose response models;
- better understanding of the role of different wastewater treatment processes for virus aggregation, inactivation and physical removal. This includes pond systems, high rate processes, filtration and disinfection processes
- improved models for representing the bioaccumulation and depuration of pathogens by shellfish given time varying concentration of pathogens in the marine waters;

As these needs are satisfied and the methodology matures, further research needs will continue to become apparent. However, we consider the power of the methodology is such that this investment in ongoing research and development is warranted and will continue to lead to a better understanding of public health risk management associated with discharges containing pathogens.

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Water Research – use of the base graphic for Figure 5.

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GLOSSARY

AdV: Adenovirus

ADF: Average Day Flow

AFRI: Acute febrile respiratory illness

DNA: Deoxyribonucleic acid

Genotype: A representation of an organism's genetic makeup.

Gi; Gastrointestinal

GI: Genotype I

GII: Genotype II

HAdV: Human Adenovirus

$HAdV_4$: Human Adenovirus of Serotype 4

HRT: Hydraulic retention time. The average time that a unit of volume is retained within a treatment unit.

Log_{10}: Logarithm to base 10. A measure of the factors of 10 included in a number.

mWs/cm^2: milli-watt.seconds per square centimeter . The widely accepted unit of measure of UV dose rate. Also stated as mJ/cm^2.

NLV: Norwalk like viruses. Includes Norovirus and caliciviruses

NoV: Norovirus

QMRA: Quantitative Microbial Risk Assessment

Ri: Respiratory

RNA: Ribonucleic acid

RT-qPCR: Reverse Transcription quantitative Polymerase Chain Reaction. A laboratory methodology for enumerating viral RNA or DNA particles. It identifies the RNA/DNA but does not differentiate between the viable (culturable) and non-viable particles.

Serotype: Usually used to describe a strain of bacteria or virus that produces a specific type of antigen. Variations within a sub-species.

UV: Ultraviolet disinfection system for water.

Virion: An entire virus particle, consisting of an outer protein shell and an inner core of nucleic acid (either RNA or DNA).

WWTP: Wastewater Treatment Plant

APPENDIX A – DOSE RESPONSE MODEL PARAMETERS

Table 2: Dose Response Models

Pathogen	*Norovirus*	*Adenovirus*	Reference
Infectivity	High	High	McBride et al. (2005)
Model Type	Beta-Binomial conditional	Binomial conditional	McBride (2005)
Dose Response Model	$Pr_{inf}(d) = 1-[B(\alpha,\beta+i)/B(\alpha,\beta)]$, where B is the standard beta function	$Pr_{inf}(d) = 1-(1-r)^i$	
Individuals per Exposure	Multiple (100)	Multiple (100)	
Parameters			
r =	NA	0.4172	
$ID_{50} = 0.693/r$	NA	1.7	Infectious dose to 50% of population (Haas et al. 2009)
α =	0.04*	NA	* Models were also run using boot strap data with 1000 pairs of (α,β) values (provided by Dr peter Teunis, RIVM, Netherlands) to account for the uncertainty in these values.
β =	0.055*	NA	
$ID_{50} = \beta*(2^{1/\alpha}-1)$	26		Infectious dose to 50% of population (Haas et al. 2009)
Harmonization	0.054	NA	McBride et al. (2013) Accounts for difference between Teunis et al. (2008) RT-PCR method and ESR RT-PCR method.
Respiratory HAdV	NA	Min 1% Most Likely 3% Max 10%	Accounting for possible presence of HAdV genotypes 2, 3 & 4 relative to the entire measured population of HAdV. (Fong et al. 2010)
Susceptibility of population (Se^+)	70%	100%	Soller et al. (2010).

Probability of Illness	0.5	0.5	Among the susceptible proportion of the population (Se^+ group), the probability of developing an illness, given that infection has occurred.

Not all the population is susceptible to Norovirus. Some appear immune[11] and so an allowance was made for this, in the model, prior to calculating the risk of infection. So, of the 100 individuals exposed during each model iteration (day at the beach) approximately 30% were discounted even before the dose response model was applied.

Among the people who are susceptible to highly infectious viruses such as Norovirus and Rotavirus, some people are more susceptible than others. The established dose response models take account of this with two-parameter distributions. However, if we only model one person with each iteration of the model, then the susceptibility of only one individual is being tested each visit and, as such, the wide variety of possible illness and infection outcomes in the population at large is not measured. Thus, we normally always model multiple individuals, in this case 100, at each site and therefore, for each model iteration. In this case however, we are effectively modelling 500 beach goers (4 beach sites plus a surf site) plus 100 shellfish eaters on each day that is modelled. This concept is considered in detail in McBride (2005a&b).

Probability of infection, following the dose response model, was converted, using a binomial function, to 1 or 0 to represent infection, or not, respectively i.e., either you are infected, or you are not.

For those who were infected, a binomial function again was used to determine if illness had resulted, in accordance with the accepted probabilities of illness, given infection has occurred. For NoV GI & GII and HAdV[2,3,4], the probabilities of illness were 0.50[12],[13] (in both cases) times the probability of infection. Again the determination of illness, or not, was determined using a binomial function and the probability of illness conditional upon infection having occurred.

[11] Teunis et al. (2008).
[12] Thebault et al. (2009).
[13] Soller et al. (2010, Table 1).