

# **ROTORUA WASTEWATER TREATMENT PLANT APPLICATIONS FOR RESOURCE CONSENTS AND ASSESSMENT OF ENVIRONMENTAL EFFECTS**

## **SUPPORT DOCUMENT NO. 6**







# ROTORUA WASTEWATER TREATMENT PLANT DISCHARGE

## PUBLIC HEALTH ASSESSMENT

PREPARED FOR ROTORUA LAKES COUNCIL

April 2018

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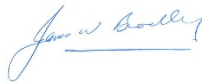
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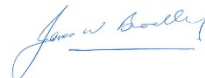
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# Executive Summary

## Background

Rotorua Lakes Council (Council) is currently preparing a resource consent application for the discharge of treated wastewater to the Black Stream channel (a geothermal stream channel), which discharges into Lake Rotorua at Sulphur Point. Based on the investigations and consultation to date, Council's preferred treatment process for the upgraded Rotorua Wastewater Treatment Plant (WWTP) is a full membrane bioreactor (MBR) treatment plant with UV disinfection and additional phosphorus removal (hereafter referred to as the Proposed Treatment Scheme).

The Assessment of Effects on the Environment (AEE) document includes eight support documents, of which this Report is Support Document No. 7.

## Council's Aim: To Be An 'Industry Leader'

In addition to the overall objectives of the consenting project, the Council's aim, with respect to public health and the ability to reuse treated wastewater, is to be an 'industry leader' by treating the wastewater to a sufficient level that it:

1. could theoretically be used directly (i.e. without dilution) for contact recreational purposes such as swimming
2. could meet the unrestricted reuse standard in relevant international guidelines<sup>1</sup>.

As a step towards this, the Council have engaged MWH to help them understand the level of treatment required to achieve the two desired outcomes. The Council also wish to further understand how this can be achieved with the Proposed Treatment Scheme.

## Expected MBR Performance

MWH have carried out a literature review to determine the likely range of expected virus reduction through the proposed MBR treatment process with a membrane nominal pore size of 0.04 micron. It is expected that the proposed MBR treatment process will effectively remove larger pathogenic micro-organisms (protozoa and bacteria), which are orders of magnitude larger than the membrane pore size.

The average log reduction expected under "typical" influent concentrations is 5.0 to 5.5 for adenovirus and 4.5 to 5.0 for enterovirus and norovirus. A higher average log reduction is expected under "outbreak" (i.e. higher) influent concentrations.

## Suitability For Contact Recreation

A Quantitative Microbial Risk Assessment (QMRA) was carried out by Graham McBride of NIWA on a range of scenarios to determine the individual's illness risk associated with the direct use of treated wastewater discharged for primary contact recreation. The QMRA is included as Appendix A to this Report.

The individual's illness risk associated with **gastrointestinal illness** resulting from direct use of the treated wastewater discharge for contact recreation:

- is less than <1% for enterovirus with 2.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 4 log reduction through the Proposed Treatment Scheme for '**typical**' influent virus concentrations

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<sup>1</sup> Unrestricted reuse does not extend to use as a potable water supply. International guidelines have been used as there is not currently a New Zealand guideline that specifies unrestricted reuse wastewater quality for viruses.

- is less than <1% for enterovirus with 4.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 5 log reduction for '**outbreak**' influent virus concentrations.

The individual's illness risk associated with **respiratory illness** (i.e. adenovirus) resulting from the direct use of the treated wastewater discharge for contact recreation:

- is less than 0.3% with 3.1 log reduction through the Proposed Treatment Scheme for '**typical**' influent virus concentrations
- is less than 0.3% with 5 log reduction through the Proposed Treatment Scheme for '**outbreak**' influent virus concentrations.

Based on the expected virus Log Reduction Value (LRV) through the proposed MBR treatment process and the QMRA results, the individual's illness risk associated with the treated wastewater discharge is expected to be less than 1% for gastrointestinal illness and 0.3% for respiratory illness. That is the treated wastewater from the proposed MBR treatment process alone is expected to meet the Council's first desired outcome (i.e. it can be used directly for contact recreation) without any additional virus reduction that may be provided by the remainder of the Proposed Treatment Scheme.

## Suitability For Unrestricted Use

The expected median treated wastewater quality from the proposed MBR treatment process was estimated from the expected average log reduction through MBR and the median influent virus concentrations used for the QMRA. The expected median treated wastewater concentration (5 enterovirus per 50 L) was then compared against the unrestricted reuse standard in international guidelines ( $\leq 2$  enterovirus per 50 L).

Based on this analysis, the proposed MBR treatment process alone would not provide a sufficient level of treatment for unrestricted reuse of the treated wastewater. UV disinfection would be required to reduce the concentration of enterovirus by at least 0.4 log.

To provide a perspective on UV disinfection requirements, a UV disinfection system sized<sup>2</sup> to provide a validated UV dose of approximately 40 mWs/cm<sup>2</sup> (based on an organism sensitivity of 12 mWs/cm<sup>2</sup>) would be expected to provide a 2.3 validated log reduction in MS2 bacteriophage and a 3 validated log reduction in enterovirus and norovirus. A UV disinfection system of this size would be in the order of \$600,000 for UV equipment supply only and \$40,000/year for power demand.

## Summary

- The proposed MBR treatment process alone is expected to satisfy the Council's first desired outcome (i.e. treated wastewater can be used directly for primary contact recreation).
- The proposed MBR treatment process with UV disinfection is expected to satisfy the Council's second desired outcome (i.e. meet the unrestricted reuse standard).
- The Proposed Treatment Scheme in its entirety includes MBR and UV which provides a multiple barrier treatment approach to pathogenic micro-organism removal.

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<sup>2</sup> For flows up to 825 L/s and treated wastewater having a UV transmittance of 65%.

## Abbreviations

Abbreviation	Definition
ADV	Adenovirus
AEE	Assessment of Effects on the Environment
<i>E. coli</i>	<i>Escherichia coli</i>
EV	Enterovirus
HAV	Hepatitis A Virus
HCGI	<b>Highly Credible Gastrointestinal Illness</b>
HRT	Hydraulic Retention Time
Log	Logarithm
LRV	Log Reduction Value
MAC	Microbiological Assessment Category
MBR	Membrane Bioreactor
Micron	Micrometer (µm)
MLSS	Mixed Liquor Suspended Solids
NTU	Nephelometric Turbidity Units
NV	Norovirus
PCR	Polymerase Chain Reaction
POSSI	Possible Gastrointestinal Illness
QMRA	Quantitative Microbial Risk Assessment
TMP	Transmembrane Pressure
UV	Ultraviolet
WWTP	Wastewater Treatment Plant

# Rotorua Lakes Council

## Public Health Assessment

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## APPENDICES

Appendix A	Quantitative Microbiological Risk Assessment For Treated Rotorua Wastewater
Appendix B	Analysis of Quantitative Microbiological Risk Assessment Inputs and Results



# 1. Introduction

## 1.1 Background

Rotorua Lakes Council (Council) is currently preparing a resource consent application for the discharge of treated wastewater to the Black Stream channel (a geothermal stream channel), which discharges into Lake Rotorua at Sulphur Point. Based on the investigations and consultation to date, Council's preferred treatment process for the upgraded Rotorua Wastewater Treatment Plant (WWTP) is a full membrane bioreactor<sup>3</sup> (MBR) treatment plant with UV disinfection and additional phosphorus removal (hereafter referred to as the Proposed Treatment Scheme).

The Assessment of Effects on the Environment (AEE) document includes eight support documents, of which this Report is Support Document No. 7.

In addition to the overall objectives of the consenting project, the Council's aim, with respect to public health and the ability to reuse treated wastewater, is to be an 'industry leader' by treating the wastewater to a sufficient level that it:

- could theoretically be used directly (i.e. without dilution) for contact recreational purposes such as swimming<sup>4</sup>.
- could meet the unrestricted reuse standard in relevant international guidelines<sup>5</sup>.

As a step towards achieving this aim, the Council want to understand the level of treatment required to achieve the two desired outcomes (i.e. minimise potential public health risks and meet the unrestricted reuse standard), which may require different levels of treatment. The Council also wish to further understand how this can be achieved with the proposed MBR and UV disinfection treatment process of the Proposed Treatment Scheme. The Council has engaged MWH to assist with these two aspects.

## 1.2 Purpose

The purpose of this Report is to consider the public health risks to recreational water users associated with the treated wastewater discharge, to estimate the treated wastewater quality, and to consider the requirements of the Proposed Treatment Scheme to mitigate any potential public health risks and meet the unrestricted reuse standard following UV irradiation and prior to any land contact.

## 1.3 Scope

MWH has carried out the following tasks.

- Reviewed recent literature of MBR performance with respect to virus reduction and, based on this review, defined a likely range of expected MBR performance at Rotorua.
- Undertaken a quantitative microbiological risk assessment (QMRA) for a range of scenarios to determine the individual illness risk associated with the direct use of treated wastewater for contact recreational purposes (i.e. swimming).
- Identified appropriate treatment requirements of the Proposed Treatment Scheme to mitigate potential public health risks based on the QMRA results and to meet the unrestricted reuse standard in

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<sup>3</sup> A combination of a membrane process with a suspended growth bioreactor.

<sup>4</sup> The public health risk associated with secondary contact recreation (such as boating) would be lower as there would be a lower ingestion rate as well as dilution within the environment prior to an individual's exposure.

<sup>5</sup> Unrestricted reuse does not extend to use as a potable water supply. International guidelines have been used as there is not currently a New Zealand guideline that specifies unrestricted reuse wastewater quality for viruses. An assumed unrestricted reuse standard of less than 2 enterovirus per 50L has been adopted for this Report.

the treated wastewater prior to any land contact arrangement or cascade discharge structure into Black Stream channel.

## 1.4 Structure

This Report is presented in five further sections:

- Section 2:** overviews waterborne micro-organisms and summarises the waterborne human pathogens that are focus of this report
- Section 3:** summarises a literature review of virus reduction through a MBR treatment process and, based on this summary, presents a likely range of expected MBR performance at Rotorua.
- Section 4:** overviews relevant guidelines, summarises the QMRA undertaken by Graham McBride of NIWA for a range of scenarios, compares the QMRA results to equivalent levels of calculated risk in New Zealand guidelines, and identifies the required virus reduction through the Proposed Treatment Scheme. This Section also provides expected treated wastewater virus concentrations from the proposed MBR treatment process at Rotorua.
- Section 5:** summarises any additional treatment requirements for the Proposed Treatment Scheme, over and above the proposed MBR treatment process, to achieve the Council's two desired outcomes (i.e. minimise potential public health risks associated with direct use for contact recreation and meet the unrestricted reuse standard).
- Section 6:** presents the conclusions from the QMRA as well as appropriate treatment requirements of the Proposed Treatment Scheme to achieve the Council's two desired outcomes based on current guidelines.

The Quantitative Microbiological Risk Assessment (QMRA) report prepared by Graham McBride of NIWA<sup>6</sup> is provided in Appendix A. Some of the analysis carried out by MWH on the QMRA inputs and results is included in Appendix B.

## 1.5 Quantitative Microbial Risk Assessment

A Quantitative Microbial Risk Assessment (QMRA) was carried out by Graham McBride of NIWA on a range of scenarios to determine the individual's illness risk associated with the direct use of treated wastewater discharged from the Rotorua wastewater treatment plant for primary contact recreational purposes (ie swimming)<sup>7</sup>.

The QMRA report (McBride, 2017), reproduced in full in Appendix A, includes:

- A discussion on QMRA methodology and inputs, including pathogens of concern, individual's exposure to pathogens and individual's likelihood of illness (i.e. dose-response). This is summarised in Section 4.3.1 of this Report.
- A tabulated summary of individual's illness risk for a range of scenarios (i.e. typical and outbreak influent concentrations, different viruses, and seven theoretical levels of virus removal through the Proposed Treatment Scheme).

MWH used the results of the QMRA to determine the minimum virus reduction required through the Proposed Treatment Scheme.

To understand if the proposed MBR treatment process alone would meet the Council's two desired outcomes based on current guidelines, MWH compared:

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<sup>6</sup> McBride, G., 2017. Quantitative microbial risk assessment for treated Rotorua wastewater. A report prepared for MWH on behalf of Rotorua District Council.

<sup>7</sup> The public health risk associated with secondary contact recreation (such as boating) would be lower as there would be a lower ingestion rate as well as dilution within the environment prior to an individual's exposure.

- The expected virus reduction through the proposed MBR with the minimum virus reduction required to be achieved through the Proposed Treatment Scheme for the individual's illness risk to be less than 1% for gastrointestinal illness and 0.3% for acute febrile respiratory illness.
- The expected concentration of viruses in the treated wastewater from the proposed MBR with the unrestricted reuse standard.

If the proposed MBR treatment process alone was shown to not meet both of the Council's desired outcomes, MWH then determined the minimum additional virus reduction required to be achieved by the remainder of the Proposed Treatment Scheme and outlined how this additional reduction may be achieved (e.g. UV disinfection, which is also part of the Proposed Treatment Scheme).

## 2. Waterborne Micro-organisms

This section presents an overview of waterborne micro-organisms that have been used to quantify the level of treatment provided at WWTPs, the individual illness risk associated with treated wastewater discharges, or both.

There are a wide variety of micro-organisms present in wastewater. Some are 'indicator' organisms, which themselves may not be pathogenic (ie cause disease) but are often used as a marker to indicate microbial contamination as they are relatively easy and cost-effective to reliably measure. Others are pathogenic and can cause a range of infections of varying severity, but can be more difficult and more expensive to measure. Key groups of micro-organisms are summarised in Table 2-1.

Table 2-1: Range of Micro-organisms<sup>8</sup>

Type	Micro-organism	Size (micron)	Comment
Pathogenic	Protozoa	3 - 14	Single cell organisms (eg Cryptosporidium and Giardia). Able to survive outside their host under adverse conditions as cysts or oocysts
	Bacteria	0.6 - 1.2 (diameter) 2 - 3 (length)	Able to multiply in dependent of host given suitable conditions. Enteropathogenic bacteria eg cholera and salmonella species. Opportunistic bacteria eg pseudomonas and streptococcus
	Virus	enterovirus: 0.018 – 0.027 adenovirus: 0.07 - 0.09 rotavirus: 0.06 – 0.08 norovirus G1 and GII: 0.02 – 0.04 hepatitis A virus: 0.027 – 0.028	Require a host (animals or people) to reproduce. Enteric viruses include enteroviruses and norovirus (both linked with gastrointestinal illness), adenoviruses (linked with respiratory illness) and hepatitis A virus. Types of viruses found in wastewater depend on those circulating in the community. Seasonal (and geographic) disease trends are seen with some viruses (eg rotavirus) but trends in virus presence in wastewater aren't as clear. Infected individuals can excrete large numbers of potentially infectious enteric viruses such as norovirus many weeks after they have recovered from illness.
Indicator	Bacteria	about 1 micron	Able to multiply independent of host given suitable conditions. Common indicator bacteria are total coliforms, including faecal coliforms (eg E. coli).
	Bacteriophage	MS2: 0.02 - 0.025 FRNA bacteriophage: 0.01 to 0.1	Viruses that infect specific bacteria and often used as process indicators for enteric virus removal or inactivation. A coliphage is a type of bacteriophage that infects E. coli. One type is F-specific phage or male-specific phage (ie MS2 coliphage) and is considered to have similar characteristics to hepatitis A virus and poliovirus (Hai, 2014). Another type is somatic phage (eg T4 coliphage) and considered to have similar characteristics to adenoviruses, reoviruses, rotaviruses and coronaviruses (Hai, 2014).

<sup>8</sup> Hai, 2014; Branch, 2015; Yin, 2012



The relative size of different micro-organisms compared to the nominal filter size proposed at Rotorua WWTP (0.04 micron) is illustrated in Figure 2-1. Figure 2-2 is the diagram used by the Rotorua Project Steering Committee in their deliberations about the treatment options and indication of the Proposed Scheme.

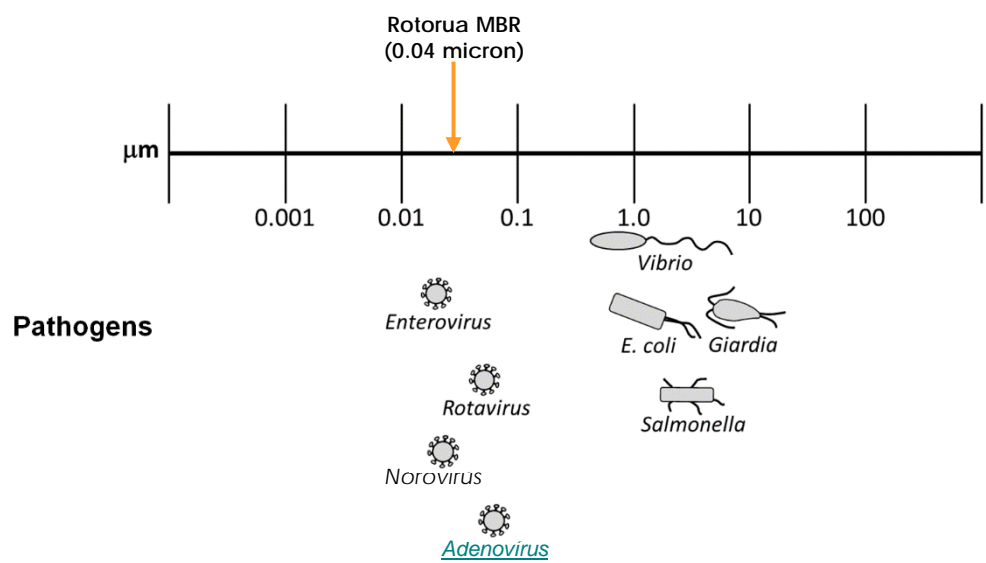


Figure 2-1: Rotorua MBR Filter Size Compared with Micro-organisms (adapted from Hai, 2014)

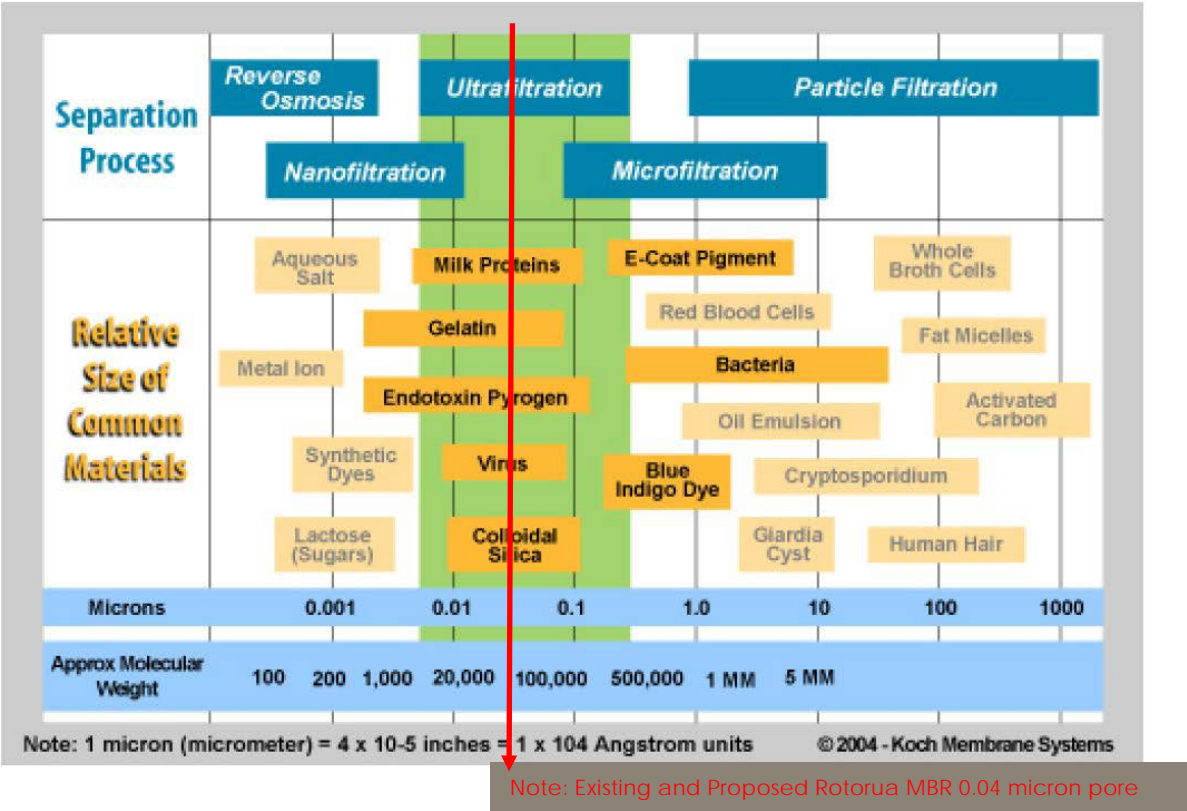


Figure 2-2: Relative Particle Size and Wastewater Treatment Process<sup>10</sup>

Human viral pathogens are focus of this Report. This is based on the expectation that a MBR-based wastewater treatment plant will effectively remove the larger pathogenic micro-organisms (protozoa and

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<sup>10</sup> From Koch Membrane Systems, 2004.

bacteria), which are orders of magnitude larger than the nominal filter size (0.04 micron) proposed at Rotorua WWTP<sup>11</sup> (McBride, 2017).

For the literature review of virus reduction via MBR, publications that provide virus data have been considered as far as possible in preference to publications that use surrogate-virus micro-organisms (eg bacteriophages like MS2 coliphage and T4 coliphage). The primary reason for this is that the removal and transport of surrogates do not necessarily correlate to those of enteric viruses in wastewater systems (Hai, 2014). For some aspects of MBR performance there is limited if any virus data available and so surrogate-virus micro-organism data have been used to provide a perspective on likely performance.

The rationale for the selection of specific human viruses used in the QMRA is outlined in Section 4.3.1 and in the QMRA report in Appendix A.

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<sup>11</sup> GE Zeeweed 500D hollow fibre units or similar (Mott MacDonald MBR Preliminary Design)

## 3. MBR Performance

### 3.1 Literature Review

This section summarises findings of a literature review of virus reduction through a MBR treatment process.

A combination of the following mechanisms contribute towards the removal of pathogens through MBR:

- size exclusion (by the membrane)
- adsorption (to the membrane or biomass) and
- biological predation (or inactivation).

The properties of the specific pathogen will determine the most dominant removal mechanism. (Branch, 2015).

For viruses larger than the membrane pore size, **size exclusion** is the predominant mechanism for removal provided the membrane integrity is sound (Branch, 2015). The nominal pore size of the MBR membrane at Rotorua WWTP will be 0.04 micron, which is smaller than the typical size of adenovirus (about 0.07 to 0.09 microns) and rotavirus (0.06 to 0.08 microns). However, it is larger than the typical size of norovirus (0.02 to 0.04 microns) and enterovirus (0.02 to 0.03 microns). Hence, based purely on relative size to the membrane pore size, norovirus and enterovirus would be expected to pass through a clean membrane.

For viruses in the order of or smaller than the membrane pore size, virus removal has been observed to be greater than that expected with a clean membrane. This enhanced virus removal is reported to be due to the **dynamic fouling layer** that builds up on the membrane and a **tendency to adsorb suspended solids**. The dynamic fouling layer on the membrane comprises organic fouling and chemical scaling (or caking), which physically reduces the effective pore size of the membrane and also entraps virus particles on the membrane surface. Virus particles will also adsorb onto the surfaces of suspended solids in the bioreactor, which essentially increases the size of the virus. (Branch, 2015; Hai, 2014; Wu, 2010).

Table 3-1 summarises the **range of human virus Log Reduction Values (LRV)** through MBRs reported in literature, along with any key points of note about the data. The focus of this review (and Table 3-1) is published papers about human virus removal (rather than other micro-organisms) using full-scale MBRs with a nominal pore size similar to that proposed at Rotorua (0.04 micron, shown in bold type in and Table 3-1) and 'real' wastewater. Due to the relatively limited number of papers based on a pore size of 0.04 micron, papers based on a larger pore size (up to 0.45 micron) have also been included in Table 3-1. The abbreviation 'avg' has been used for 'average' in the table.

Table 3-1: Observed Virus LRV Through MBR12

Virus	LRV	Comment	Reference
Adenovirus	4.1-5.6, avg 5.0	Full-scale MBR, <b>0.04 micron</b> (nominal). 8 month study period, 8 samples. Average LRV of overall adenovirus was $5.0 \pm 0.6$ across MBR. Adenovirus concentrations of 1000 – 10000 gc/L detected in MBR effluent. Study also looked at three adenovirus species. Average LRV were $4.1 \pm 0.9$ (A species), $4.6 \pm 0.5$ (C species), $6.5 \pm 1.3$ (F species).	Kuo, 2010 (also cited in Hirani, 2014; Hai, 2014; Yin, 2012; Simmons, 2011 <sup>13</sup> )
Adenovirus	3.9 – 5.5	Full-scale MBR, <b>0.04 micron</b> , 10 year old membranes. 6 month sampling period. Adenovirus detected in 6/6 influent samples and 14/17 permeate samples. Samples include those taken after CIP. Average LRV results	Chaundhry, 2015 (also cited in

<sup>12</sup> All of the papers presented in Table 3-1 are based on virus detection or enumeration using PCR-based methods.

<sup>13</sup> Yin 2012 and Hai 2014 report Simmons (2011) as separate case study to Kuo (2010) but Simmons (2011) uses same dataset from Traverse City WWTP from Jan to Aug 2008. Kuo (2010) presents raw data as a time series whereas Simmons (2011) presents a statistical analysis with range in LRV for adenovirus of 4.1 to 6.3 (average 5.5), and so only the results from Kuo (2010) have been included in the table:

Virus	LRV	Comment	Reference
		presented in paper; individual paired LRV results not shown.	Yin, 2016)
Adenovirus	4.4 avg	Full-scale MBR, <b>0.04 micron</b> . Adenovirus detected in MBR effluent at low concentrations.	Purnell, 2016
Adenovirus	2.38 - >4.86, median >3.67	Three MBR plants, 0.4 micron. Adenovirus detected in 11/11 MBR influent samples and 5/11 permeate samples. Adenovirus in MBR effluent samples with concentration up to 19 gc/L when concentration in MBR influent varied from 220 to 180,000 gc/L	Francy, 2012 (also cited in Hirani, 2014)
Adenovirus	Virus detected in MBR effluent	Nine full-scale MBRs. 3 MBR effluent samples at each plant, influent not analysed. Adenovirus detected in most MBR effluent samples for all plants.	Hirani, 2013
Adenovirus	Virus detected in MBR effluent	Pilot-scale MBR, 0.1 micron. Adenovirus detected before and after membrane <i>chemical cleaning</i> (0.2% NaClO). Adenovirus detected before and after <i>membrane breach</i> (cut membrane, filtrate turbidity > 0.5 NTU). Influent concentration not measured. Note: enterovirus, rotavirus and hepatitis A virus not detected in the samples despite having a smaller diameter (~0.3 micron) than adenovirus (~0.06-0.09 micron). Author suggests it may be due to other viruses being present in substantially lower concentrations, although no influent data to confirm this.	Hirani, 2014
Enterovirus	4.1-6.8, avg 5.1	Full-scale MBR, <b>0.04 micron</b> (nominal). 8 month study period, 8 samples. Average LRV of enterovirus was $5.1 \pm 0.9$ across MBR. Enterovirus concentrations of 10 – 100 gc/L detected in MBR effluent.	Simmons, 2011 (also cited in Yin, 2012; Hai, 2014; Sano, 2016)
Enterovirus	>1.79 avg	Full-scale MBR, 0.4 micron. Enterovirus detected in 18/23 influent samples and 5/17 permeate samples, with a mean concentration of 160 gc/L. Log removal value of $1.79 \pm 0.55$ , where detection limit used if not detected in permeate.	Ottoson, 2006 (also cited in Sano, 2016; Hirani, 2014)
Enterovirus	>2.2 - 4.74, median >3.40	Three MBR plants, 0.4 micron. Enterovirus detected in 10/11 MBR influent samples and 2/11 permeate samples. Enterovirus detected in MBR effluent samples with concentration of 5.3 gc/L when concentration in MBR influent varied from 240 to 290,000 gc/L	Francy, 2012 (also cited in Hirani, 2014)
Enterovirus	>0.3 - >3.2	Full-scale MBR, 0.4 micron. 16 month study period. Enterovirus detected in 13/19 MBR influent samples and 4/19 permeate samples (2/4 of these were when MLSS concentration was reduced to 50-60% of normal operation level). When enterovirus was detected in MBR effluent, mean LRV was $1.6 \pm 0.4$ .	Miura, 2015 (also cited in Sano, 2016)
Enterovirus	Virus not detected in MBR effluent	Nine full-scale MBRs. 3 MBR effluent samples at each plant, influent not analysed).	Hirani, 2013
Enterovirus	Virus not detected in MBR effluent	Pilot-scale MBR, 0.1 micron. Enterovirus not detected before and after membrane <i>chemical cleaning</i> (0.2% NaClO). Enterovirus not detected before and after <i>membrane breach</i> (cut membrane, filtrate turbidity > 0.5 NTU). Influent concentration not measured.	Hirani, 2014
Hepatitis A virus	Virus not detected in	Nine full-scale MBRs. 3 MBR effluent samples at each plant, influent not analysed.	Hirani, 2013



Virus	LRV	Comment	Reference
	MBR effluent		
Hepatitis A virus	Virus not detected in MBR effluent	Pilot-scale MBR, 0.1 micron. Hepatitis A virus (HAV) not detected before and after membrane <i>chemical cleaning</i> (0.2% NaClO). HAV not detected before and after <i>membrane breach</i> (cut membrane, filtrate turbidity > 0.5 NTU). Influent concentration not measured.	Hirani, 2014
Norovirus	2.3 avg	Full-scale MBR, <b>0.04 micron</b> . Norovirus GI and GII detected in MBR effluent in low concentrations. Results of GI and GII combined to demonstrate removal of norovirus as a whole. This study also showed similar concentrations of norovirus GI and GII in influent but higher concentrations of adenovirus. Higher LRV observed for adenovirus (average of 4.4; see other entry in this table).	Purnell, 2016
Norovirus	>1.14 avg	Full-scale MBR, 0.4 micron. Norovirus detected in 8/22 influent samples and 3/17 permeate samples, with a mean concentration of 22 gc/L. Log removal value of $1.14 \pm 0.88$ , where detection limit used if not detected in permeate. Genogroup not specified.	Ottoson, 2006
Norovirus	3.3 – >6.8	Full-scale MBR, 0.45 micron. Norovirus GI and GII detected in MBR effluent. Results of GI and GII combined. Lower LRVs of 0.9 and 1.3 observed during period of membrane integrity issues. Norovirus GI detected in 14/15 MBR influent samples and 14/32 permeate samples. Norovirus GII detected in 15/15 MBR influent samples and 10/32 permeate samples. Overall norovirus GI and GII were always close to the detection limit. Higher removals observed when higher concentrations in MBR influent.	Sima, 2011 (also cited in Sano, 2016)
Norovirus GI	>0* - >5.5 *nondetectable in influent	Full-scale MBR, 0.4 micron (nominal). Sampling during winter (December – April) when norovirus concentrations expected to be higher. GI detected in 8/11 influent samples and 2/11 effluent samples. Minimum LRV was about 1 when GI was detected in influent and effluent.	da Silva, 2007 (also cited in Hai, 2014; Kuo, 2010 and Yin, 2012)
Norovirus GI	>1.51 – 3.32, median >3.02	Three MBR plants, 0.4 micron. Norovirus GI detected in 7/11 MBR influent samples and 2/11 permeate samples. Norovirus detected in MBR effluent samples with concentration of about 11 gc/L when concentration in MBR influent varied from about 50 to 20,000 gc/L	Francy, 2012
Norovirus GII	>3.5 - >4.8, avg >3.9 Virus not detected in MBR effluent	Full-scale MBR, <b>0.04 micron</b> (nominal). 8 month study period, 8 samples. Average LRV of norovirus GII was $3.9 \pm 0.5$ across MBR. Note: Norovirus GII detected in 4/8 MBR influent samples and 0/8 MBR effluent samples. LRV range based on influent samples where norovirus detected and test detection limit for norovirus. This study also showed lower concentrations of norovirus GII than adenovirus and enterovirus in MBR influent and lower LRV. Norovirus GI was not detected in influent. Higher LRV observed for adenovirus and enterovirus (average of 5.5 and 5.1, respectively; see other entries in this table).	Simmons, 2011 (also cited in Yin, 2012; Sano, 2016)
Norovirus GII	4.6 – 5.7	Full-scale MBR, <b>0.04 micron</b> , 10 year old membranes. 6 month sampling period. Norovirus detected in 4/4 influent samples and 14/17 permeate samples. Samples	Chaundhry, 2015 (also cited in

Virus	LRV	Comment	Reference
		include those taken after CIP. Average LRV results presented in paper; individual paired LRV results not shown.	Yin, 2016 and Sano, 2016)
Norovirus GII	>2.2 - >5.2 Virus not detected in MBR effluent	Full-scale MBR, 0.4 micron (nominal). Sampling during winter when norovirus concentrations expected to be higher. GII detected in 12/12 influent samples and 0/8 effluent samples	da Silva, 2007 (also cited in Hai, 2014; Kuo, 2010 and Yin, 2012)
Norovirus GII	>0.2 - >3.4	Full-scale MBR, 0.4 micron. 16 month study period. Norovirus detected in 18/19 MBR influent samples and 5/19 permeate samples (3/5 of these were when <i>MLSS concentration was reduced to 50-60% of normal operation level</i> ). When norovirus was detected in MBR effluent, mean LRV was $1.3 \pm 0.8$ .	Miura, 2015 (also cited in Sano, 2016)
Rotavirus	Virus not detected in MBR effluent	Nine full-scale MBRs. 3 MBR effluent samples at each plant, influent not analysed.	Hirani, 2013
Rotavirus	Virus not detected in MBR effluent	Pilot-scale MBR, 0.1 micron. Rotavirus not detected before and after membrane <i>chemical cleaning</i> (0.2% NaClO). Rotavirus not detected before and after <i>membrane breach</i> (cut membrane, filtrate turbidity > 0.5 NTU). Influent concentration not measured.	Hirani, 2014

Overall, studies of MBR performance to date indicate high removal of viruses is achieved in many cases but suggest MBRs are not able to serve as an absolute barrier against viruses (Yin, 2016).

The key observations from the papers summarised in Table 3-1 are:

- increased virus LRV is generally observed with a smaller membrane pore size
- increased virus LRV is generally observed with increased influent concentration
- increased virus LRV is generally observed for adenovirus compared to enterovirus or norovirus when influent concentrations are similar. This is expected based on virus size relative to membrane nominal pore size; adenovirus is in the order of twice the pore size, whereas enterovirus and norovirus are between 0.5 to 1.0 times pore size
- in some cases virus LRV is underestimated due to virus not being detected in the MBR effluent (or MBR influent)
- with a pore size of 0.04 micron and sufficiently high 'typical' MBR influent concentrations, the minimum mean LRV is expected to be 5.0 to 5.5 for adenovirus and 4.5 to 5.0 for enterovirus and norovirus. Higher LRV would be expected with higher MBR influent concentrations.

The virus LRV reported in literature varied between studies. There are several possible reasons for this (Branch, 2015; Rames, 2016), including the following.

- Influent virus concentrations. The higher the initial influent virus concentration, the higher the possible LRV that can be observed.
- Pre-treatment of wastewater (ie primary treated wastewater or untreated wastewater). Virus particles adsorb to solids, and so primary treated wastewater may contain a lower concentration or an altered ratio of virus species. Note Kuo (2010) observed no removal of human adenovirus by primary sedimentation.
- Use of 'real' wastewater (ie from a wastewater treatment plant) or 'synthetic' wastewater (eg manufactured with similar chemical composition or de-ionised water inoculated with virus particles).

- Use of seeded versus indigenous micro-organisms. In some cases wastewater is 'innoculated' to artificially increase the concentration of virus particles in the influent. Seed micro-organisms can behave differently to indigenous micro-organisms.
- Use of other micro-organisms such as coliphages of a similar size to viruses. Ottoson, 2006 (and cited in Hai, 2014) observed a marked variation in the removal of tested indicator organisms, with bacterial indicators more efficiently removed than coliphages, which were more efficiently removed than enterovirus and norvirus genomes.
- Different experimental setup – i.e. bench-scale, pilot-scale and full-scale MBR plants, use of acclimatised biomass, different pore size, different operating regimes.
- Differences in test methods used to quantitate influent and treated wastewater virus concentrations. Culture-based methods report the quantity of virus that are able to infect a given host whereas Polymerase Chain Reaction (PCR) based methods report the number of virus genome copies<sup>14</sup> detected, which cannot distinguish between infectious and non-infectious viruses. PCR-based methods often report concentrations that are several orders of magnitude higher than those determined from infectivity assays (Chaudhry, 2015) and probably underestimate infectious virion removal (Ottoson, 2006). PCR-based methods also use a variety of PCR primers, which often have different levels of specificity (ie detect a particular type of virus, say adenovirus, or detect a particular virus strain). In both cases, the methods used to collect and prepare a sample for analysis as well as the test detection limit may result in variation between laboratories.
- Some review papers have not included the LRV for a paired set of influent and MBR permeate samples<sup>15</sup> if there were no virus particles detected in the treated wastewater, which will underestimate the observed range of LRV.

### 3.1.1 Operational Variability

The papers presented above focus on typical LRV that can be expected during normal operation. This section provides some perspective on impact of membrane cleaning, membrane breach (i.e. loss of membrane integrity) and changes in operational parameters on LRV.

Several studies have been carried out to better understand the impact (both extent and duration) of hydraulic and chemical cleaning on LRV. Many are based on bench-scale MBR and larger membrane pore sizes (ie >0.04 micron) but generally suggest for full-scale MBR plants there is minimal impact in LRV and that any impact is short-lived. Papers of note include the following.

- Yin (2016) observed that hydraulic cleaning (both pressure relaxation and permeate backwash) with a 0.45 micron bench-scale MBR led to about a 1 log reduction in removal of human adenovirus, however removals returned to pre-cleaning levels within 16 hours after backwash. Jacangelo (1995) (as cited in Hai, 2014) observed virus removal by MF/UF membranes increased with time with accumulation of foulants but that it did not decrease after hydraulic backwash.
- Hirani (2014) observed that enterovirus, rotavirus and hepatitis A virus were not detected in MBR effluent before or after chemical cleaning with hypochlorite. Adenovirus were detected in both situations but the author suggests it may be due to substantially higher influent concentrations, however there was no influent data to confirm this.
- van den Akker, 2014 observed that chemical cleaning (soaking and aerating membranes in hypochlorite and caustic, chemical backwash in citric acid and then hypochlorite/caustic soda) of full-scale MBR reduced the LRV of *E. coli* and total coliforms each by about 1 log but did not affect the removal of bacteriophage or clostridia. Viruses were not measured. The observations were attributed

<sup>14</sup> A unit of measure commonly used when reporting virus concentrations obtained from PCR-based methods

<sup>15</sup> A paired set is the term used when one influent sample and one MBR permeate sample is taken at the same time, sometimes adjusted for hydraulic retention time.

to the fact that these indicators absorb well to larger flocs, which are readily rejected by a clean membrane. The LRV improved over the 5 day period following cleaning.

For full-scale MBRs, the removal of viruses (and other micro-organisms) is dependent on the integrity of the MBR system. A loss of integrity due to the membrane (eg abnormally large pores, compromised glue line, holes) or filtration system (eg compromised o-rings, broken mechanical seals) can result in a spike in both turbidity and microorganisms in the treated wastewater (Hai, 2014; Branch, 2015). Under breached conditions, the filtrate typically increases immediately after relaxation/backwash and gradually reduces to a previously observed value once the membrane plugs with activated sludge after a few minutes of filtration (Zha, 2008 as cited in Hai 2014). In a full-scale MBR study, Hirani (2014) observed that enterovirus, rotavirus and hepatitis A virus were not detected in MBR effluent before or after a membrane breach. Adenovirus were detected in both situations but the author suggests it may be due to substantially higher influent concentrations, however there was no influent data to confirm this.

Currently it is unknown if general decay or decomposition of the membrane may result in decreased rejection over the long-term operation (Hai, 2014).

The Australian Water Recycling Centre of Excellence funded a project to develop a national validation framework for MBRs in Australia. As part of this project, data was analysed from 11 full scale MBRs to better understand impact of **operational parameters** on LRV for virus-surrogates (somatic coliphage, FRNA bacteriophage), bacteria (*E. coli*) and protozoan (*C.perfringens*). The MBRs range in pore size 0.04 to 0.4 micron (most 0.04 micron, as is proposed for the full scale Rotorua WWTP upgrade and is presently used in the existing MBR that treats one third of the flow). This analysis showed that there was a higher likelihood of low LRV with low Hydraulic Retention Time (HRT), high flux, high permeability, low Transmembrane Pressure (TMP), high permeate turbidity, low Mixed Liquor Suspended Solids (MLSS) and high dissolved oxygen (Branch, 2015). Further work is needed to confirm whether or not these findings correlate to impact on LRV for human viruses.

### 3.1.2 Rotorua Monitoring Data

Currently at the Rotorua WWTP, two thirds of the flow is treated by an activated sludge-based plant and the remainder is treated by a MBR-based plant. To get an understanding of the current level of virus reduction through the existing MBR-based plant, limited virus monitoring was carried out in 2016 under typical operating conditions. Samples were taken from the untreated wastewater and from the MBR permeate (i.e. the MBR treated wastewater). The monitoring results are summarised in Table 3-2.

Table 3-2: Virus Monitoring at Current Rotorua WWTP

Sampling Date	Adenovirus (genome copies/L)		Norovirus Genotype I (genome copies/L)		Norovirus Genotype II (genome copies/L)	
	Influent	Permeate	Influent	Permeate	Influent	Permeate
22/8/16	980,000	not detected	130,000	<25 (TBC)	6,800,000	<25
30/8/16	8,800,000	not detected	18,000	<25	7,300,000	720
5/9/16	1,600,000	not detected	29,000	<25	11,000,000	90

The results show that viruses are typically reduced by the following:

- adenovirus – more than 5.3 log removal<sup>16</sup> (LRV limited by influent concentration)
- norovirus GI – more than 2.9 log removal (LRV limited by influent concentration)
- norovirus GII – at least 4.0 log removal.

<sup>16</sup> A detection limit of 5 genome copies per L was assumed to calculate log reduction in adenovirus



## 3.2 Expected MBR performance

Table 3-3 provides the likely range of expected MBR performance at Rotorua (0.04 micron) with respect to virus reduction for the following two scenarios:

- **“typical”** influent virus concentrations, considered to be representative for the majority of the time
- **“outbreak”** influent virus concentration, considered to be representative following a disease outbreak in the community. Characterised by a short spike in virus concentration at the start of the outbreak followed by a decline to “typical” concentrations.

Table 3-3: Expected Virus LRV Through Proposed MBR Treatment Process (0.04 micron)

Influent Scenario	Adenovirus	Enterovirus	Norovirus
“typical”	5.0 to 5.5	4.5 to 5.0	same as enterovirus
“outbreak” <sup>17</sup>	6.2 to 6.7	5.7 to 6.2	6.5 to 7.0

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<sup>17</sup> Expected virus LRV for “outbreak” scenario based on influent concentrations (and LRV) being 1.2 log higher than “typical” scenario for adenovirus and enterovirus and being 2.0 log higher for norovirus.

## 4. Quantitative Microbiological Risk Assessment

### 4.1 Overview

A quantitative microbiological risk assessment (QMRA) was carried out to determine the potential risk of infection associated with the direct use of treated wastewater discharged from the Rotorua wastewater treatment plant for primary contact recreational purposes (ie swimming)<sup>18</sup>.

The QMRA has been undertaken in recognition of the requirements of the microbiological guidelines for receiving water inherent with the *“Microbiological Water Quality Guidelines for Marine and Freshwater Recreational Areas”* published by the Ministry for the Environment and the Ministry of Health (2003).

This section outlines the relevant guidelines, summarises the QMRA undertaken by Graham McBride of NIWA<sup>19</sup> for a range of scenarios, compares the QMRA results to equivalent levels of calculated risk in New Zealand guidelines, and identifies the required virus reduction through the Proposed Treatment Scheme. The complete QMRA report prepared by Graham McBride of NIWA is presented in Appendix A.

This section then provides an overview of expected treated wastewater quality from the proposed MBR treatment process based on the influent virus concentration used for the QMRA and the expected virus LRV through the proposed MBR. This is to enable the Council to understand if the proposed MBR treatment process alone would meet the Council’s two desired outcomes or if additional virus is reduction is required to be achieved by the remainder of the Proposed Treatment Scheme (e.g. UV disinfection).

### 4.2 QMRA Guidelines

#### 4.2.1 Background

It is recognised that the definition of an acceptable level of risk of symptomatic infection is a difficult choice. In considering the calculated risk of infection, it is important to recognise the risk levels inherent in the existing bathing water guidelines and those applied to other areas within related industries.

#### 4.2.2 Microbiological Water Quality Guidelines for Marine and Freshwater Recreational Areas

The Ministry for the Environment originally published the Microbiological Water Quality Guidelines for Marine and Freshwater Recreational Areas (the “Guidelines”) in June 2002. An updated version of the Guidelines was completed in June 2003. The Guidelines replace the previous Ministry for the Environment/Ministry of Health Recreational Water Quality Guidelines published in November 1999.

The Guidelines were developed over an extensive period of consultation with regional and local councils and health authorities, and present a preferred approach to monitoring recreational waters. It should also be noted that they are not legislated standards that must be adhered to at all times. Furthermore, the Guidelines state that they **“should not be directly applied to assess microbiological quality of water that is impacted by a nearby point source discharge of treated wastewater without first confirming that they are appropriate. This is particularly important for disinfected effluent<sup>20</sup>...”**.

The Guidelines use a combination of qualitative risk grading of the catchment, supported by the direct measurement of appropriate faecal indicators to assess the suitability of a site for recreation. In addition, alert and action guideline levels are used for surveillance throughout the bathing season. The two components to providing a grading for an individual site are:

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<sup>18</sup> The public health risk associated with secondary contact recreation (such as boating) would be lower as there would be a lower ingestion rate as well as dilution within the environment prior to an individual’s exposure.

<sup>19</sup> McBride, G., 2017. Quantitative microbial risk assessment for treated Rotorua wastewater. A report prepared for MWH on behalf of Rotorua Lakes Council.

<sup>20</sup> Where disinfection is used to reduce the density of indicator bacteria in treated wastewater, the presumed relationship between *E. coli* and pathogen presence may be altered. In waters, receiving such treated wastewater, *E. coli* may not provide an accurate estimate of the risk of infection.

- The *Sanitary Inspection Category (SIC)*, which generates a measure of the susceptibility of a water body to faecal contamination
- Historical microbiological results, which generate a *Microbiological Assessment Category (MAC)* and provides a measure of the actual water quality over time.

The two criteria provide an overall Suitability for Recreation Grade (SFRG), which describes the general condition of a site at any given time, based on both risk and indicator bacteria counts. This grade provides the basis for telling people whether or not the water is suitable for recreational use from a public health perspective.

The Microbiological Assessment Category (MAC) system for marine waters<sup>21</sup> is considered the most appropriate to use when interpreting the QMRA results for the Rotorua WWTP discharge as it is based on the individual illness risk related to viruses. The system for freshwater<sup>22</sup> is based on the individual infection risk associated with campylobacter, which will be retained on the membrane as campylobacter are about a hundred times larger in size than the membrane pore size of the proposed MBR.

The Guidelines provide a classification (i.e. a Suitability for Recreation Grade) based on a number of contact recreation associated illness risk thresholds. The Guidelines classify waters as “very good” for its Suitability for Recreation Grade as posing a less than 1% risk of gastrointestinal illness (GI) and less than 0.3% risk for acute febrile respiratory illness (AFRI); “good” grading as posing 1% to 5% risk of GI (and between 0.3% and 1.9% risk for AFRI). For waters that are classified as in a “fair” condition, the illness risk thresholds are between 5% to 10% and 1.9% to 3.9% for GI and AFRI respectively. Waters that pose an illness risk of greater than 10% for GI and greater than 3.9% for AFRI are classified as being in a “poor” condition.

### 4.2.3 Historical Water Quality Guidelines: Local and International

Prior to the publication of the 2003 Guidelines, the Recreational Water Quality Guidelines published by the Ministry for the Environment/Ministry of Health in November 1999 served as a basis for evaluating the suitability of water bodies for recreational contact. These guidelines were based on guidelines implemented by the USEPA following the work of Dufour (freshwater) and Cabelli (marine water). The “acceptable” illness (not infection) rates associated with the development of these studies were as follows:

- a bathing season illness rate of 8 cases of HCGI<sup>23</sup> per 1,000 swimmers (i.e. 0.8 percent) as inherent in the development of a number of historical guidelines for freshwater bathing water
- a bathing season illness rate of 19 cases of HCGI per 1,000 swimmers (i.e. 1.9 percent) as inherent in the development of a number of historical guidelines for marine bathing water.

It should however be noted the “acceptable” illness rates stated above were not chosen a priori but calculated following the decision to accept the risk level associated with the standard at the time.

### 4.2.4 Marine Bathing Beach Survey

It is of interest to note that the Bathing Beach Study conducted in New Zealand<sup>24</sup> for marine bathing beaches reported the following.

- A baseline illness rate of 58 cases per 1,000 individuals for people attending the beach but not entering the water. The baseline illness rate comprised 17 per 1,000 incidences associated with highly credible gastrointestinal illness (HCGI), 18 per 1,000 incidences of possible gastrointestinal illness (POSSI)<sup>25</sup>, and 23 per 1,000 incidences of respiratory illness.

<sup>21</sup> the marine guidelines are based on incidence of illness from epidemiological studies, and hence incorporate the effect of morbidity (ie the rate of infections becoming illnesses)

<sup>22</sup> the freshwater guidelines are based on incidence of infection derived from a quantitative risk assessment

<sup>23</sup> HCGI = High credible gastrointestinal illness; ANYGI= any gastrointestinal illness.

<sup>24</sup> McBride GB, Salmond CE, Bandaranayake DE, Turner SJ, Lewis GD and Till DG (1998), “Health Effects of Marine Bathing in New Zealand”, *Int. J Environ Health Res.*, 8: 173-189..

<sup>25</sup> Possible gastrointestinal illness (POSSI) is equal to any gastrointestinal illness (ANYGI) minus highly credible gastrointestinal illness (HCGI)

- An illness rate of 56 per 1,000 paddlers, i.e. waders who entered the water but did not immerse the head. The paddler illness rates comprised 21, 6 and 29 incidences per 1,000 individuals for HCGI, POSSGI and respiratory illness respectively.
- An illness rate of 98 per 1,000 swimmers, i.e. people who entered the water and immersed the head. The swimmer illness rates comprised 21, 38 and 39 incidences per 1,000 individuals for HCGI, POSSGI and respiratory illness respectively.

## 4.3 QMRA

### 4.3.1 QMRA Approach

A QMRA was carried out by Graham McBride of NIWA on a range of scenarios to determine the individual illness risk associated with the direct use of treated wastewater discharged from the Rotorua WWTP for contact recreation only<sup>26</sup>. The methodology used to carry out the QMRA is detailed in Appendix A. This section provides a summary of the general approach used for Rotorua WWTP.

The approach used to carry out the QMRA comprised four basic steps:

1. Select the pathogens of concern, based on water-related diseases that may arise (i.e. gastrointestinal illness and respiratory illness) and have been commonly used to define water quality standards. The pathogens chosen for contact recreation were:
  - Adenovirus (linked with respiratory diseases) – It is very infective and low and may be present in treated wastewater.
  - Enteroviruses (linked with gastroenteritis) – It is less infective, but health consequences can be more severe than adenovirus
  - Norovirus<sup>27</sup> (linked with gastroenteritis) – There is increasing evidence of its prevalence in treated wastewater and dose-response relationships are now available.
2. Assess exposures to the pathogens, based on typical and outbreak influent virus concentration (taken from other New Zealand studies), range of theoretical virus log reduction values for a future Rotorua WWTP (i.e 2 log to 8 log reduction), direct use of treated wastewater (i.e. no dilution in the receiving waters) and primary contact recreation (i.e. swimming).
3. Characterise the pathogen's dose-response, based on published studies and data from viral illness outbreaks and infection.
4. Calculate the health risks, using Monte Carlo statistical modelling to reflect the likely variations in a range of assumptions that define the treated wastewater quality, an individual's exposure and an individual's risk of infection and illness. A random sample is taken from each distribution to calculate an individual's risk of illness. The sampling procedure is repeated many times to simulate a large population being exposed to water that may, on some occasions, be contaminated.

Key assumptions used to carry out the QMRA, in addition to those outlined above, include:

- The Proposed Treatment Scheme for Rotorua is appropriately designed and operated to accommodate current and future flows and loads
- there are no WWTP bypasses around treatment processes
- there is no dilution or decay of residual pathogenic micro-organisms in the discharge treated wastewater once discharged into the water environment.

<sup>26</sup> The scope of the QMRA summarised in this Report was agreed with Rotorua Lakes Council

<sup>27</sup> Dose-relationship for norovirus genotype GI has been used in this QMRA.



### 4.3.2 Individual Illness Risk

This section of the report summarises the results of the QMRA, which is presented in Appendix A. Data are presented in this section to illustrate the individual's illness risk for a range of scenarios in comparison to equivalent risk levels in New Zealand guidelines. Based on this comparison, the required virus reduction through the Proposed Treatment Scheme in its entirety (i.e. MBR, UV disinfection and phosphorus removal) for the individual's illness risk to be less than the risk levels associated with a "very good" grading is identified.

Figure 4-1 presents the individual's illness risk associated with contact recreation based on the selected pathogens of concern (i.e viruses) and a range of theoretical virus log reductions through the Proposed Treatment Scheme in its entirety. The results have been grouped based on the following:

- **Influent virus concentration.** *Typical* virus concentrations are considered to be representative for the majority of the time. *Outbreak* virus concentrations are considered to be representative following a disease outbreak in the community, which is characterised by a short spike in virus concentration at the start of the outbreak followed by a decline to "typical" concentrations
- **Type of infection and illness.** *Respiratory* illness, based on adenovirus. *Gastrointestinal* illness, based on enterovirus and norovirus (aggregated and disaggregated). As outlined in Section 4.2.2, the MAC system in the Guidelines comprises a four-tiered scale with different risk cut-offs for the mean individual illness (respiratory and gastrointestinal) risk. In Figure 4-1, MAC A is shaded green, MAC B blue, MAC C yellow and MAC D orange.

The individual gastrointestinal illness risks for norovirus have been analysed based on an infection model that assumes the virus is either all aggregated (ie all present in clusters of more than one virion) or all disaggregated (ie all separate virions). If norovirus is present in treated wastewater from the Proposed Treatment Scheme, it is more likely it is present in the disaggregated form than in the aggregated form. The main reason for this is that for norovirus to be present in the treated wastewater, it needs to have passed through the proposed MBR membrane. A disaggregated norovirus particle (about 0.02 to 0.04 micron) is similar in size to the proposed membrane pore size (0.04 micron), whereas an aggregated norovirus particle will generally be larger in size.

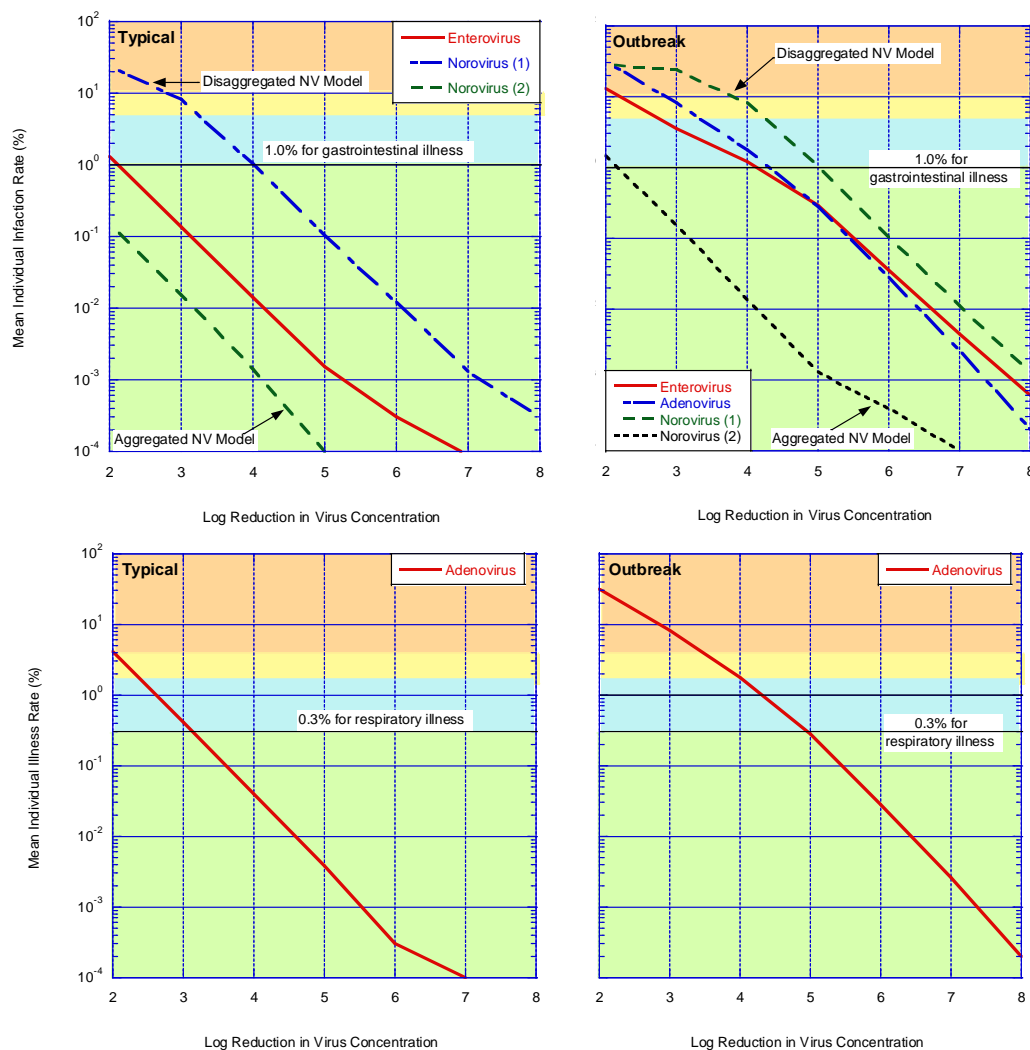


Figure 4-1: Individual's Illness Rate (%) for Gastrointestinal illness (upper graphs) and Respiratory Illness (lower graphs) Associated with Primary Contact Recreation (Swimming) based on Range of Virus Log Reduction Through Proposed Treatment Scheme for Typical and Outbreak Influent Virus Concentrations of Adenovirus, Enterovirus, Norovirus (aggregated and disaggregated)

Note: MAC colour code: MAC A green, MAC B blue, MAC C yellow and MAC D orange.

The QMRA with respect to **gastrointestinal** illness for the Proposed Treatment Scheme has shown that:

- the individual's illness risk is less than 1% for enterovirus with 2.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 4 log reduction<sup>28</sup> through the Proposed Treatment Scheme for '**typical**' influent virus concentrations
- the individual's illness risk is less than 1% for enterovirus with 4.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 5 log reduction for '**outbreak**' influent virus concentrations.

<sup>28</sup> The required log reduction is less for norovirus (aggregated) than for norovirus (disaggregated). The mean individual gastrointestinal illness risk is less than 1% for aggregated norovirus with less than 2 log reduction through the Proposed Treatment Scheme. However, if norovirus is present in MBR treated wastewater, it is more likely to be present in the disaggregated form and so the disaggregated results are of most importance from a public health perspective.

The QMRA with respect to **respiratory** illness for the Proposed Treatment Scheme has shown that:

- the individual's illness risk is less than 0.3% for adenovirus with 3.1 log reduction through the Proposed Treatment Scheme for '**typical**' influent virus concentrations
- the individual's illness risk is less than 0.3% for adenovirus with 5 log reduction through the Proposed Treatment Scheme for '**outbreak**' influent virus concentrations.

The required virus log reduction through the Proposed Treatment Scheme for the individual's illness risk to be less than 1% and 0.3%, respectively, for gastrointestinal and respiratory illnesses is summarised in Table 4-1.

Table 4-1: Required Virus LRV Through Proposed Treatment Scheme For Individual's Illness Risk to be Less Than Required Levels

Influent Scenario	Gastrointestinal Illness (<1%)		Respiratory Illness (<0.3%)
	Enterovirus	Norovirus	Adenovirus
"typical"	2.1	4.0	3.1
"outbreak" <sup>29</sup>	4.1	5.0	5.0

In some cases, the above log reductions may overly conservative or unnecessarily high. The following should be considered when interpreting the results:

- The "typical" influent concentration and relative proportion of different viruses can change over the year and between WWTPs. For example at Napier WWTP, influent norovirus concentrations remain high whereas influent enterovirus and adenovirus concentrations tend to be lower and much more **variable** (McBride, 2016)<sup>30</sup>. Whereas different trends have been seen elsewhere (Purnell, 2016; Ottoson, 2006; Miura, 2015). The probability distribution of influent concentrations used for the QMRA is given in Appendix B.
- The influent concentration distribution used for the 'outbreak' scenario (see Appendix B) mimics **substantially greater enterovirus and adenovirus** concentrations observed for some weeks in Mangere WWTP influent in a Scoping Study in May – July 1999. Unusually, the concentrations for both viruses in the Scoping Study were up to 1,000 times larger than in other investigations at the time and virus concentrations as high as these have not been observed during the regular ongoing monitoring at Mangere since (McBride, 2016).
- The QMRA considered two forms of norovirus – aggregated and disaggregated, with the same influent concentration assumed for both forms (ie all disaggregated or all aggregated). **Aggregation of noroviruses** markedly reduces norovirus illness risk when those viruses are present in low concentrations (McBride, 2014a in Clarks Beach MRA), which can be seen as a 2 to 3 log difference in individual illness risk when comparing aggregated and disaggregated norovirus results for the same LRV. As noted previously, if norovirus is present in treated wastewater from the Proposed Treatment Scheme, it is **more likely it is present in the disaggregated form** than in the aggregated form. The main reason for this is that for norovirus to be present in the treated wastewater, it needs to have passed through the proposed MBR membrane and a disaggregated norovirus particle (about 0.02 to 0.04 micron) is similar in size to the proposed membrane pore size (0.04 micron). A precautionary approach has been taken when interpreting the QMRA results by assuming that all the noroviruses are disaggregated.

<sup>29</sup> Expected virus LRV for the "outbreak" scenario are based on influent concentrations (and LRV) being 1.2 log higher than "typical" scenario for adenovirus and enterovirus and being 2.0 log higher for norovirus.

<sup>30</sup> Hirani (2014) reported one study with similar influent concentrations of enterovirus and adenovirus in the order of 100 to 100,000 gc/L and another with influent concentrations of adenovirus in the order of 1,000,000 to 10,000,000 gc/L.

- The QMRA considered the use of treated wastewater directly for contact recreation. The treated wastewater is proposed to discharge to the Black Stream channel (a geothermal channel), which discharges into Lake Rotorua at Sulphur. This means that, in reality, there will at least some **dilution** within the receiving environment prior to an individual being exposed to the discharge. To provide a perspective on this, a 10-fold dilution equates to a 1 log reduction and a 100-fold dilution equates to 2-log reduction. In reality there will also be at least some **decay** within the receiving environment prior to an individual being exposed. The Black Stream channel is a geothermal channel, with a low pH and a high temperature, but the effects of such an environment on virus reduction has not been considered in the QMRA.
- In comparison to primary contact recreation, a lower virus log reduction through the MBR would be required to achieve the same level of risk associated with **secondary contact** recreation due to the lower ingestion rate and dilution within the receiving environment.

## 4.4 Expected Treated Wastewater Quality from Proposed MBR

The expected median concentration of enterovirus in the treated wastewater discharged from the proposed MBR treatment process is presented in Table 4-2. This treated wastewater quality has been estimated from the expected virus LRV through the proposed MBR (see Table 3-3, lower end of range used) and the median influent virus concentrations used for the QMRA (see Appendix A).

Table 4-2: Estimated Median Influent and Treated Wastewater Concentrations of Enterovirus

Influent Scenario	Influent	Assumed LRV Through MBR	Treated Wastewater
"typical"	3,000 number/L	4.5	0.09 number/L 5 number/50L
"outbreak"	50,000 number/L	5.7	0.10 number/L 5 number/50L

The expected treated wastewater concentration presented in Table 4-2 will be used in Section 5 to determine if the proposed MBR treatment process alone would meet the Council's two desired outcomes or if additional virus reduction is required to be achieved by the remainder of the Proposed Treatment Scheme (e.g. UV disinfection).

## 5. Additional Requirements

### 5.1 Ability of Proposed MBR to Meet Council's Aim

The Council's aim, in respect to public health protection and the ability to reuse treated wastewater, is to be an 'industry leader' by treating the wastewater to a sufficient level that it:

1. could theoretically be used directly (ie without dilution) for contact recreational purposes such as swimming. This would require the mean individual illness risks associated with this activity to be less than 1% for gastrointestinal illness and 0.3% for acute febrile respiratory illness.
2. Could meet the unrestricted reuse standard in relevant international guidelines.

Based on the expected virus log reduction through the proposed MBR treatment process (see Table 3-3) and the results of the QMRA (see Table 4-1), the individual's illness risk associated with the treated wastewater discharge is expected to be less than the required levels (i.e. <1% for gastrointestinal illness and <0.3% for respiratory illness). This demonstrates that the treated wastewater from the proposed MBR treatment process alone is expected to meet the Council's first desired outcome (i.e. it can be used directly for contact recreation) without additional virus reduction provided by the remainder of the Proposed Treatment Scheme.

There is no New Zealand guideline and only a few guidelines available internationally that provide a numeric value for an acceptable concentration of human enteric viruses in treated wastewater that is reused without restriction (ie unrestricted reuse). Guidelines that are available from New South Wales, Australia, provide a limit for enterovirus of  $\leq 2$  virus per 50 L for recycled water for urban reuse<sup>31</sup>. No values are available for adenovirus or norovirus. Based on this limit and estimated treated wastewater quality from the proposed MBR treatment process in Table 4-2 the proposed MBR treatment process alone would not provide a sufficient level of treatment for unrestricted reuse of the treated wastewater. Additional virus reduction would be required to be achieved by the remainder of the Proposed Treatment Scheme (e.g. UV disinfection) to meet the Council's second treatment requirement (ie meet unrestricted reuse standard).

### 5.2 Additional Virus Log Reduction Value and Treatment Options

Based on the expected MBR performance (Section 3.2), the QMRA results (Section 4.3.2) and the expected treated wastewater quality from the proposed MBR treatment process (see Section 4.4), the following additional virus reduction is required to be achieved by the remainder of the Proposed Treatment Scheme to meet the Council's aim and desired outcomes:

- 0.4 log reduction in enterovirus to meet the unrestricted reuse guidelines to reduce the enterovirus concentration from 5 per 50L to 2 per 50L. It is noted that no guideline value is available for adenovirus or norovirus.

Additional virus reduction is not required for the individual's illness risk to be lower than for 1% for gastrointestinal illness and 0.3% for acute febrile respiratory illness.

The additional virus reduction requirements and broad treatment option(s) to achieve additional virus reduction are summarised in Table 5-1. Broad treatment options have been provided in Table 5-1 for all pathogens of concern to provide a perspective, as unrestricted reuse guideline values may become available for other viruses in the future.

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<sup>31</sup> In 1989 guidelines were developed for viruses in recycled water for urban reuse in New South Wales (NSW). The guidelines were developed by a committee involving the NSW Department of Health and the NSW Department of Public Works. It is understood that the proposed guideline values were not established from specific scientific data. The guidelines were strongly influenced by other guidelines / standards (i.e. World Health Organisation: zero per 10 L of drinking water. and in Arizona, USA: 1 per 40 L for unrestricted reuse.



Table 5-1: Residual Virus Reduction Requirements and Broad Options

Pathogen of Concern	Required LRV for Contact Recreation	Required LRV for Reuse	Possible Treatment Option	Comments
Adenovirus	nil	no guideline value	Chlorine disinfection	Readily inactivated by chlorine disinfection. Significantly more resistant to UV disinfection.
Enterovirus	nil	0.4	UV disinfection	Readily inactivated by UV disinfection. More resistant to chlorine disinfection.
Norovirus	nil	no guideline value		

## 5.3 UV Disinfection

The treated wastewater from the proposed MBR treatment process requires UV disinfection enable it to meet the unrestricted reuse standard of 2 enterovirus per 50L<sup>32</sup>. UV disinfection would also provide further reduction in norovirus and, to a much lesser extent, adenovirus. It would also provide redundancy in the treatment process, should there be any occasion where there is reduced virus removal through the MBR system.

To provide a perspective on UV disinfection requirements, a UV disinfection system has been sized to provide a validated UV dose of approximately 40 mWs/cm<sup>2</sup>, based on an organism sensitivity of 12 mWs/cm<sup>2</sup>, flows up to 825 L/s, and treated wastewater having a UV transmittance of 65%. Such a UV disinfection system would be expected to provide a 2.3 validated log reduction in MS2 bacteriophage and a 3 validated log reduction in enterovirus and norovirus.

The indicative capital costs for a UV disinfection system of this size (UV equipment supply only), excluding gst, are in the order of \$600,000<sup>33</sup>, with indicative operating costs (power demand only) in the order of \$40,000 per year<sup>34</sup>.

## 5.4 Further Mitigation Measures

If the Council wish to provide further log reduction in concentration of adenoviruses under typical conditions or provide security on MBR performance, chlorine disinfection is likely to be more cost effective than UV disinfection. Adenovirus is significantly more resistant to UV disinfection than enterovirus and norovirus but is readily inactivated by chlorine disinfection.

If free chlorine is utilised as a disinfectant, a free chlorine CT of less than 1 mg.min/L has been shown to achieve 2 log removal of adenoviruses (Jacangelo et al, 2002 as cited in Hirani, 2014). A chlorine disinfection system has not been sized or costed at this stage as neither the QMRA or the expected treated wastewater quality suggest that it would be needed.

<sup>32</sup> A reduction of at least 0.4 log in enterovirus is required to meet the unrestricted reuse standard.

<sup>33</sup> This cost is based on a single open channel system with 6 modules of lamps (duty) with 1 module as standby. This cost excludes costs associated with investigation, design, consenting, associated works (ie civil, mechanical, electrical and controls), construction and commissioning,

<sup>34</sup> Based on 6 modules of lamps (duty) at 50%, 365 day/year operation, 8c per kWh.

## 6. Conclusions

Key conclusions from this Report are summarised in this section.

### Expected MBR Performance

Based on a literature review, MWH identified the likely range of expected MBR performance at Rotorua (0.04 micron). The average log reduction expected under "typical" influent concentrations is 5.0 to 5.5 for adenovirus and 4.5 to 5.0 for enterovirus and norovirus. A higher average log reduction is expected under "outbreak" (i.e. higher) influent concentrations.

### Suitability For Contact Recreation Purposes

A QMRA was carried out on a range of scenarios to determine the individual's illness risk associated with the direct use of treated wastewater discharged from the Proposed Treatment Scheme for primary contact recreation.

The individual's illness risk associated with **gastrointestinal illness** resulting from direct use of the treated wastewater discharge for contact recreation:

- is less than 1% for enterovirus with 2.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 4 log reduction for '**typical**' influent virus concentrations
- is less than 1% for enterovirus with 4.1 log reduction through the Proposed Treatment Scheme and for norovirus (disaggregated) with 5 log reduction for '**outbreak**' influent virus concentrations.

The individual's illness risk associated with **respiratory illness** resulting from the direct use of the treated wastewater discharge for contact recreation:

- is less than 0.3% for adenovirus with 3.1 log reduction through the Proposed Treatment Scheme for '**typical**' influent virus concentrations
- is less than 0.3% for adenovirus with 5 log reduction through the Proposed Treatment Scheme for '**outbreak**' influent virus concentrations.

Based on the expected virus LRV through the proposed MBR treatment process and the QMRA results, the individual's illness risk associated with the treated wastewater discharge is expected to be less than desired (i.e. <1% for gastrointestinal illness and <0.3% for respiratory illness). That is the treated wastewater from the proposed MBR treatment process alone is expected to meet the Council's first desired outcome (i.e. it can be used directly for contact recreation) without additional virus reduction provided by the remainder of the Proposed Treatment Scheme.

### Suitability for Unrestricted Reuse

Based on an unrestricted reuse standard of  $\leq 2$  enterovirus per 50 L and the estimated treated wastewater quality from the proposed MBR treatment process of 5 enterovirus per 50 L, the proposed MBR treatment process alone would not provide a sufficient level of treatment for unrestricted reuse of the treated wastewater. UV disinfection would be required to reduce the concentration of enterovirus by at least 0.4 log.

To provide a perspective on UV disinfection requirements, a UV disinfection system sized<sup>35</sup> to provide a validated UV dose of approximately 40 mWs/cm<sup>2</sup> (based on an organism sensitivity of 12 mWs/cm<sup>2</sup>) would be expected to provide a 2.3 validated log reduction in MS2 bacteriophage and a 3 validated log reduction in enterovirus and norovirus. A UV disinfection system of this size would be in the order of \$600,000 for UV equipment supply only and \$40,000/year for power demand.

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<sup>35</sup> For flows up to 825 L/s and treated wastewater having a UV transmittance of 65%.

## Summary

In summary:

- the proposed MBR treatment process alone is expected to satisfy the Council's first desired outcome (ie treated wastewater can be used directly for primary contact recreation)
- the proposed MBR treatment process with UV disinfection is expected to satisfy the Council's second desired outcome (ie meet the unrestricted reuse standard)
- the Proposed Treatment Scheme in its entirety includes MBR and UV which provides a multiple barrier treatment approach to pathogenic micro-organism removal.

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# Appendices



# Appendix A   Quantitative Microbiological Risk Assessment For Treated Rotorua Wastewater

# Quantitative Microbial Risk Assessment for Treated Rotorua Wastewater

*MWH Ltd*

*March 2017*

Prepared by:  
Graham McBride


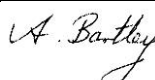

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## Executive summary

Rotorua District Council is examining various options to improve treatment of wastewater at its Wastewater Treatment Plant, including several levels of advanced secondary treatment and disinfection. In so doing it has taken the view that human health risks should be calculated for these levels for the treated effluent *before* any mixing occurs with the receiving (lake) water. Those risks are calculated using QMRA techniques (Quantitative Microbial Risk Assessment), as explained hereafter.

This report explains the choice of pathogens to be used for the QMRA. In situations like these, direct assessment of pathogens is called for, since national guidelines counsel against reliance on faecal indicator bacteria. Three viral pathogens have been selected: enterovirus, norovirus and adenovirus.

The treatment levels assumed cover six orders-of-magnitude. That is, efficacies of virus removals (comparing wastewater influent and effluent virus concentrations) are taken as 2, 3,..., 8 (e.g., dividing the pathogen influent concentrations by a factor up to  $10^8$ ).

The results of this assessment procedure are expressed for each viral pathogen in terms of Individual Illness Risk. These results will be utilised in a forthcoming report by MWH Ltd.

# 1 Background

Rotorua District Council is examining various options for improved treatment of its Wastewater Treatment Plant, including several levels of advanced secondary treatment and disinfection. In so doing it has taken the view that human health risks arising from contact with the effluent should be calculated for these levels for the treated effluent *before* any mixing occurs with the receiving (lake) water. Those risks are calculated using Quantitative Microbial Risk Assessment (QMRA) techniques, as explained hereafter.

Accordingly, this report explains the choice of pathogens to be used for the QMRA: in situations like these, direct assessment of pathogens is called for, since national guidelines counsel against reliance on faecal indicator bacteria (MfE/MoH 2003, pp. 3–4). Three viral pathogens have been selected: enterovirus, norovirus and adenovirus. Justification of these choices is presented.

The treatment levels assumed cover six orders-of-magnitude. That is, efficacies of virus removals between wastewater influent and effluent are taken as 2, 3,..., 8 (e.g., dividing the pathogen influent concentrations by a factor up to  $10^8$ ).<sup>1</sup>

Results are detailed and utilised in a forthcoming report by MWH Ltd. Accordingly, this report lacks any discussion or conclusions sections.

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<sup>1</sup> These order-of-magnitude reduction factors are often called “log removals”, it being implicit that the logarithms are to base 10.

## 2 Methods: Conducting the QMRA

Quantitative Microbial Risk Assessment (QMRA) consists of four basic steps:

1. Select the hazard(s), i.e., the pathogen(s) of concern—exposure to which can give rise to illness.
2. Assess exposures to the pathogens at key sites (in terms of pathogen concentrations and duration of exposure).
3. Characterise the pathogens' dose-response.
4. Calculate and communicate the health risks.

The “Quantitative” aspect of QMRA has particularly to do with item 4—calculating risks—in which we use Monte Carlo statistical modelling. This calls for repetitive sampling from distributions and ranges of key variable concentrations, rather than just using single average concentration values. This approach is particularly important given that the majority of the risk is caused by combinations of inputs toward the extremes of their concentration ranges, the combined effects of which may not be detected when using average concentration values.

### 2.1 Selecting the pathogens of concern

In addressing this issue, extensive use has been made of international literature and previous New Zealand studies (e.g., McBride 2011, 2012, 2015, 2016).

#### 2.1.1 Illnesses of potential concern and their pathogens

To select appropriate pathogens, we first need to consider the water-related diseases that may arise.

Many of the illnesses that may be contracted from exposure to waters contaminated by human-derived treated wastewater are not “notifiable”. Reporting for some that are<sup>2</sup> may not necessarily capture or represent much of the disease burden.<sup>3</sup> So, when considering such matters, microbiological water quality guidelines developed both in New Zealand (MfE/MoH 2003) and internationally (WHO 2003) are based on several investigations that have led to the understanding that risks associated with wastewater-contaminated water comprise two types of infection and illness: (i) Gastrointestinal disease, via ingestion during recreational water-contact, and consumption of raw shellfish flesh; (ii) Respiratory ailments, via inhalation of aerosols formed when water-skiing, surfing or by nearby breaking waves.

Other categories of diseases, especially ear, nose, throat and skin infections have generally not been included in QMRA studies to date, not least because while there is some evidence of associations between these ailments and microbial water quality (Charoenca & Fukioka 1994), dose-response models (WHO 2003, p. 55) have not been developed.

Table 2-1 lists potential waterborne diseases and their aetiological agents (i.e., pathogens), derived from the ANZECC guidelines (ANZECC & ARMICANZ 2000). It also indicates whether our assessment of the particular pathogen should be based on contact recreation or shellfish consumption exposure routes, and gives a brief rationale for this assessment.

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<sup>2</sup> See the list at <http://www.health.govt.nz/our-work/diseases-and-conditions/notifiable-diseases>

<sup>3</sup> For example, “acute gastroenteritis” is notifiable but is subject to the requirement that “not every case of acute gastroenteritis is necessarily notifiable, only those where there is a suspected common source or from a person in a high risk category (for example, a food handler, an early childhood service worker) or single cases of chemical, bacterial, or toxic food poisoning such as botulism, toxic shellfish poisoning (any type) and disease caused by verotoxin or Shiga toxin-producing *Escherichia coli*.”

**Table 2-1: Screening of treated wastewater-borne microorganisms of public health significance.**

Pathogen	Include?	Main disease caused	Rationale
<b>Bacteria</b>			
<i>Campylobacter spp.</i>	No	Gastroenteritis	Poor survival in seawater.
Pathogenic <i>E. coli</i>	No	Gastroenteritis	Low concentration expected in treated wastewater.
<i>Legionella pneumophila</i>	No	Legionnaires' disease	No evidence of environmental infection route.
<i>Leptospira sp.</i>	No	Leptospirosis	Low concentration expected in treated wastewater.
<i>Salmonella sp.</i>	No	Gastroenteritis	Low concentration expected in treated wastewater.
<i>Salmonella typhi</i>	No	Typhoid fever	Rare in New Zealand.
<i>Shigella sp.</i>	No	Dysentery	Low concentration expected in treated wastewater.
<i>Vibrio cholerae</i>	No	Cholera	Rare in New Zealand.
<i>Yersinia enterocolitica</i>	No	Gastroenteritis	Low concentration expected in treated wastewater.
<b>Helminths</b>			
<i>Ascaris lumbricoides</i>	No	Roundworm	Rare in New Zealand.
<i>Enterobius vermicularis</i>	No	Pinworm	Low concentration expected in treated wastewater.
<i>Fasciola hepatica</i>	No	Liver fluke	Rare in New Zealand.
<i>Hymenolepis nana</i>	No	Dwarf tapeworm	Rare in New Zealand.
<i>Taenia sp.</i>	No	Tapeworm	Rare in New Zealand.
<i>Trichuris trichiura</i>	No	Whipworm	Rare in New Zealand.
<b>Protozoa</b>			
<i>Balantidium coli</i>	No	Dysentery	Low concentration expected in treated wastewater.
<i>Cryptosporidium</i> oocysts	No	Gastroenteritis	Will be removed by proposed wastewater treatment processes.
<i>Entamoeba histolytica</i>	No	Amoebic dysentery	Rare in New Zealand.
<i>Giardia</i> cysts	No	Gastroenteritis	Moderate survival in seawater but will be removed by proposed wastewater treatment processes.
<b>Viruses</b>			
Adenoviruses	Yes (SW only) <sup>4</sup>	Respiratory disease <sup>5</sup>	Very infective. Significant concentrations may be present in wastewater.
Enteroviruses	Yes (SW and SF)	Gastroenteritis	Less infective, but health consequences can be more severe than for exposure to adenovirus.
Hepatitis A virus	No	Infectious hepatitis	Minimal concentration in treated wastewater; very infective. Can affect recreational water users in contaminated waters.
Noroviruses	Yes, exploratory only (SW & SF)	Gastroenteritis	Increasing evidence of its prevalence in treated wastewater. Clinical trials and dose-response now available. However, it hasn't been possible to culture in the laboratory until now. <sup>6</sup> This makes assessment of treatment efficacy problematic.
Rotavirus	No	Gastroenteritis	Limited evidence of waterborne infection in NZ; infection in children would be of concern. <sup>7</sup> Difficult to translate units used in clinical trial (Focus Forming Units, FFU, Ward et al. 1986) to those used in culture methods. See section 2.1.3 for detailed justification for its omission.

A notable feature of Table 2-1 is the selection of human viral pathogens. In general terms, for sites impacted by WWTPs processing well-treated human-derived wastewater there is widespread agreement that human viruses are the principal aetiological agent causing gastrointestinal disease among water users and consumers of raw shellfish, e.g., Lodder & de Roda Husman (2005), Sinclair et al. (2009).<sup>8</sup> Accordingly, bacteria and protozoa have been excluded from consideration in this QMRA on the expectation that an upgraded wastewater treatment plant will effectively remove these larger microbes.

### 2.1.2 Selected viruses

The relative merits of the candidate viruses (for which some form of identified dose-response curve is available) are addressed in Table 2-2, with main microbiological features summarised in Appendix A.

#### *Gastrointestinal illness*

Enteroviruses (coxsackie virus and echovirus) are selected for three reasons:

1. Their evaluation is by culture, whereas noroviruses to date have had to be analysed by qPCR methods,<sup>9</sup> and the ratio of infectious/total virus numbers can be expected to vary through the wastewater treatment process.
2. Enteroviruses can cause longer-term illnesses.
3. Clinical trial data and associated infection dose-response relationships based on culture methods are available and have already been used for the health risk assessment associated with the Manukau Wastewater Treatment Plant (DRG 2002, Simpson et al. 2003).

Noroviruses have also been included in a somewhat exploratory mode, recognising that while they are often held to be the main aetiological agent for health risk following exposure to waters containing human-derived treated wastewater residues, their enumeration poses difficulties in terms of assessing WWTP removal efficacy and subsequent infectivity (da Silva et al. 2007, Hewitt et al. 2011, Sima et al. 2011, Flannery et al. 2012, Doré et al. 2013, McBride 2014a). QMRAs based on noroviruses have been conducted elsewhere in New Zealand, e.g., Napier and New Plymouth (McBride 2011, 2012).<sup>10</sup> We assume that the removal of noroviruses through the WWTP will be at least as effective as that inferred for enteroviruses.

#### *Respiratory illness*

For this illness category we have only one choice: adenovirus. We are not aware of any other respiratory agents, appropriate to treated wastewater, for which dose-response information is available. Its merits and drawbacks are listed in Table 2-2.

<sup>4</sup> "SW" = swimming; "SF" = shellfish.

<sup>5</sup> Adenoviruses can also cause pneumonia, eye infections and gastroenteritis.

<sup>6</sup> A new culture-based method has recently been published—Jones et al. (2014): <http://www.ncbi.nlm.nih.gov/pubmed/25378626>.

<sup>7</sup> Rose & Sobsey (1993) have documented a rationale for concern about potential contamination of shellfish by rotavirus, but risk appears to have been over-estimated (they equated FFU with actual numbers of virions).

<sup>8</sup> This is not necessarily true for agricultural wastes in rural settings, where bacteria and protozoa predominate—with few exceptions (hepatitis E, some rotaviruses), animal viruses are not pathogenic to humans.

<sup>9</sup> "qPCR" refers to quantitative Polymerase Chain Reaction, a molecular laboratory test that essentially counts the number of virions in a sample, whether infectious or inactivated.

<sup>10</sup> "Norovirus" subsumes the term "Norwalk virus". The clinical trial reported and analysed by Teunis et al. (2008) was for the original Norwalk virus (genotype group GI.1)—it had been stored in a laboratory for some years. Since the time of the first identified norovirus outbreak (in Norwalk, Ohio, 1968) a number of similar caliciviruses have been identified, in genogroups I–V. Current practice is to regard the infectivity of GI.1 norovirus as equivalent to all noroviruses that affect humans (particularly GI and GII).



**Table 2-2: Comparison of the merits and limitations of viruses for which dose-response information is available.**

<b>Virus</b>	<b>Advantages</b>	<b>Disadvantages</b>
<b><u>Gastrointestinal</u></b>		
Enterovirus	Can induce more serious long-term effects compared to other viruses (Haas et al. 1999, DRG 2002, Simpson et al. 2003). Its inclusion is warranted given that it can cause more serious longer-term illnesses. <sup>11</sup>	Restricted to echovirus 12, the only enterovirus for which an infection dose-response relationship is available. Nevertheless, enterovirus by culture captures more than just echovirus, so, for example, would also capture Coxsackie virus. Meaning of "dose" not clear, giving rise to two quite different infection ID <sub>50</sub> values (54 and 1052). <sup>12</sup> See Appendix C.
Norovirus	Reported to be the most common aetiological agent in receiving waters (e.g., Sinclair et al. 2009). Infection ID <sub>50</sub> is in the order of 20 virions (among susceptible people), but the dose-response curve rises steeply from the origin, such that ~20% of people may become infected after ingestion of just one virion—see Figure B-1(b), emphasising that a precautionary approach should be taken when modelling this virus.	Efficacy of wastewater treatment in removing infectious noroviruses is difficult to establish. Restricted to Norwalk virus—norovirus genotype I.1. But note that an outbreak study (Thebault et al. 2013) identified other genotypes to be, if anything, at least as virulent. In the absence of results to the contrary, and taking an appropriate precautionary approach, noroviruses in treated wastewater are assumed to be not aggregated - were they to be aggregated, health risks would be lessened. May require a conversion from the PCR method used in the clinical trial (Lindsmith et al. 2003, Teunis et al. 2008), as described in McBride et al. (2013).
Rotavirus	Particularly affects children. The most infective virus for which published dose-response data is available. Has been used as a “model virus” in earlier QMRAs, for Warkworth (Stott & McBride 2009), Army Bay (Palliser 2011), Snell’s Beach (Palliser & Pritchard 2012).	Not as prevalent in treated wastewater as noroviruses. Doses in the one available clinical trial (Ward et al. 1986) were measured in terms of "Focus Forming Units" (FFU), with the lowest "dose" set at 0.009 FFU. So FFU numbers need to be multiplied by an unknown factor to index doses of discrete virions (see the approach taken in a USA-wide study, McBride et al. 2013). See section 2.1.3 for details.
Hepatitis A	A serious illness. Dose-response function indicates virulence (infection ID <sub>50</sub> = <2).	Present in very low numbers in treated wastewater relative to noroviruses.
Coxsackie (an enterovirus)	May particularly affect children (Suptel 1963).	Studied by Couch et al. (1965) for coxsackie A21 so restricted to respiratory illness response. Present in low numbers in treated wastewater. Dose-response function (Haas et al. 1999) indicates moderate virulence (infection ID <sub>50</sub> = 48).
<b><u>Respiratory</u></b>		
Adenovirus	Found routinely in treated wastewater (DRG 2002, Simpson et al. 2003, Thompson et al. 2003, Hewitt et al. 2011). Very resistant to disinfection (is double-stranded DNA). A common cause of gastrointestinal illness (especially the 40/41 complex). Can be applied to respiratory infections, and therefore be relevant for surfers and/or water-skiers.	Dose-response only for adenovirus 4, a respiratory aetiological agent. Haas et al. (1999) report fitting a single-parameter exponential model to data reported by Couch et al. (1966a) giving rise to an infection ID <sub>50</sub> less than 2 virions. However, most adenoviruses are not respiratory agents. Applying the adenovirus 4 dose-response model to all adenoviruses for gastrointestinal illness appears to over-estimate the dose-response for that form of illness (we can expect more substantial response of the human body's defences to gastrointestinal infection compared to respiratory infection). Applying the model to only the respiratory portion of total adenoviruses requires assumptions about their proportional presence in treated wastewater (Kundu et al. 2013). The latter authors also considered other studies by Couch et al. (1966b, 1969).

<sup>11</sup> For example, coxsackievirus type B (an enterovirus) is now recognised as the most common viral aetiological agent associated with heart disease (Haas et al. 1999).

<sup>12</sup> Infection ID<sub>50</sub> is a quantity derived from clinical trials of pathogen infectivity. It is the pathogen dose that would result in 50% of an exposed population becoming infected.

### 2.1.3 Why not select rotavirus?

As noted in Table 2-2, rotavirus has been used in other QMRA exercises (Warkworth, Army Bay, and Snell's Beach), in the period 2009–2011. In these exercises it was used as a “model virus”, representing general pathogenicity, i.e., including the likes of norovirus.<sup>13</sup>

Since that time an infection dose-response function for norovirus has been identified (and used in other places<sup>14</sup>) and a fuller understanding of the enterovirus dose-response has been gained. For such reasons these two viruses, and not rotaviruses, are now to be used both as important individual pathogens and as indicators of the possible impact of other (unknown) pathogens.

## 2.2 Assessing exposure

### 2.2.1 Predicting doses

To turn concentrations into doses we need:

1. Influent virus concentrations.
2. Treatment plant virus removal efficacy.
3. Ingestion or inhalation rates for water users.

Details on how these factors have been modelled and enumerated are given in Table 2-3.

Note that water ingestion rates by swimmers—a key component of dose-calculation—have been studied using novel biochemical procedures in a pilot study (Dufour et al. 2006). These authors report a clinical trial observing 53 volunteers involved in recreational swimming in an outdoor community swimming pool. Swimmers were assumed to ingest similar amounts of water during swimming in pools or in freshwater due to similar behaviours in each (frequently immersing their heads under the surface and remaining in the water for long periods of time). Cyanuric acid was used to trace water ingestion because it is present in outdoor swimming pools (as a decomposition product of chlorine-stabilising chloroisocyanurate) and passes through the human body unmetabolised. For each swimmer, the volume of water ingested during active swimming events lasting at least 45 minutes was calculated. It has become standard practice to apply these ingestion rates to coastal water recreation.<sup>15</sup>

More recently an analysis of Dufour's full study has been undertaken by ESR (2016): “New Zealand Exposure Factors Handbook: Recommended Values”. Client Report 16002, Prepared for Ministry of Health, 28 p.

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<sup>13</sup> In so doing, the units of dose used in developing the infection dose-response function (Focus Forming Units, see Table 2-2) has been ignored, i.e., it was assumed that FFU = numbers of rotaviruses per litre of sample. This can lead to a gross exaggeration of the risk of rotavirus illness.

<sup>14</sup> Norovirus has been used as the pathogen for QMRA studies for Westland Milk/Hokitika (Stott & McBride 2011), Napier (McBride 2011, 2016), New Plymouth (McBride 2012), Hawera/Eltham/Whareora (Palliser et al. 2013), Akaroa (McBride 2016) and Motueka (McBride 2014b).

<sup>15</sup> Personal communication: Jeff Soller, Soller Environmental, California (<http://www.sollerenvironmental.com/env/main/Home.html>).

**Table 2-3: Distributions and inputs for the QMRA.** Plain numbers in the Statistics column are for typical health conditions in the Rotorua community; italicised numbers are for the rare case that there is a norovirus illness outbreak in that community

Component	Statistics		Distributions/comments
Influent virus concentrations			Bounded “hockey stick” distribution (McBride 2005a), strongly right-skewed with a hinge at the 95%ile.
Influent enterovirus concentration, per litre	Minimum = $4 \times 10^2$	$4 \times 10^3$	Mimicking high values found for Mangere influent in a “Scoping study” in May-July 1999 (Table B1, DRG 2002, where missing values for 24 & 26 May were advised by Mr Peter Loughran, MWH, on 7 11/2003—these values are plotted on Figure 3.3.5 of the DRG report). Most usually the concentrations are 1,000–10,000 per litre (DRG 2002, Table B6). <sup>a</sup>
	Median = $3 \times 10^3$	$5 \times 10^4$	
	Maximum = $10^4$	$5 \times 10^7$	
Influent adenovirus concentration, per litre	Minimum = $10^3$	$10^4$	Rationale as above. Most usually the concentrations are 1,000–10,000 per litre (DRG 2002, Table B6): 10% of these concentrations are assumed infectious for respiratory illness effects (Kundu et al. 2013 have noted that a minority of adenovirus strains cause respiratory illness).
	Median = $3 \times 10^3$	$5 \times 10^4$	
	Maximum = $10^4$	$10^7$	
Influent norovirus concentration, genome copies per litre	Minimum = $10^3$	$10^3$	Typical range found for New Zealand cities (e.g., Napier, New Plymouth—McBride 2011, 2012, 2016).
	Median = $10^4$	$10^6$	
	Maximum = $10^6$	$10^7$	
Duration of swim (hours)	Minimum = 0.1		Child.
	Median = 0.5		
	Maximum = 4		
Swimmers water ingestion rate, mL per hour	Minimum = 20		Lognormal distribution, for a child (adult rate is half this rate). For a review on this see Wood et al. (2015, sec. 6.2.1).
	Median = 53		
	Std. Dev. = 75		
	Maximum = 270		
Dose-response equations and parameters	–		<ul style="list-style-type: none"> <li>Adenoviruses, simple binomial [eq. (4)]; <math>r = 0.4142</math> (so <math>ID_{50, \text{infection}} \approx 2</math>), <math>\text{Pr}(\text{ill} \mid \text{Infection}) = 0.5</math> (Soller et al. 2010),</li> <li>Enterovirus, beta-binomial [eq. (5)]: <math>\alpha = 1.3</math>, <math>\beta = 75</math> (so <math>ID_{50, \text{infection}} = 53</math>); <math>\text{Pr}(\text{ill} \mid \text{Infection}) = 0.4</math> (Gerba ).</li> <li>Norovirus, disaggregated: beta-binomial [eq. (5)]: <math>\alpha = 0.04</math>, <math>\beta = 0.055</math> (so <math>ID_{50, \text{infection}} = 26</math>); <math>\text{Pr}(\text{susceptible}) = P = 0.72</math> (Teunis et al. 2008); <math>\text{Pr}(\text{ill} \mid \text{Infection}) = 0.60</math> (Soller et al. 2010). Also Messner et al. (2014) exponential equation for aggregated norovirus: <math>\text{Pr}_{\text{inf}} = [1 - P e^{d/\mu}]</math>, where <math>d</math> = dose and <math>\mu</math> = mean aggregate size (taken as <math>\mu = 1106</math>).</li> </ul>

<sup>a</sup> Those high values, persisting for over a month, have not been seen in subsequent Mangere influent virus assays. Yet were they to recur during an undetected outbreak in the contributing community, one could expect elevated illness risk.

## 2.3 Characterising dose-response

These relationships are mostly inferred from data reported by “volunteer studies” (i.e., clinical trials). These have been done for a restricted number of viruses. In these studies healthy adult volunteers (typically between 50 and 100, in groups of 10 or so) are individually challenged with a pathogen dose and their infection and illness states are monitored for a few days thereafter. Occasionally data from viral illness outbreaks have been available from which dose-response information can be inferred.<sup>16</sup> Appendix B contains a full description of how these relationships are derived, and Appendix C discusses the special case of enteroviruses (via a clinical trial on echovirus). Note that in order to perform QMRA calculations, comparability between the definition of “dose” used in the clinical trial or outbreak study and the methods used in assessing virus concentrations in treated wastewater for a particular facility is required. For example, noroviruses cannot be cultured, so a quantitative molecular-based laboratory procedure (Reverse Transcription Polymerase Chain Reaction “RT-qPCR”) is used to detect the norovirus genome. Since RT-qPCR detects genetic material, the method picks up both viable and non-viable viruses. This overestimation has been accounted for in the dose-response model used in the QMRA.

## 2.4 Conducting the risk assessment

In order to adequately reflect limits to knowledge on key features of the risk assessment, Monte Carlo statistical modelling is used (Haas et al. 1999, McBride 2005a). In simpler models key inputs are described by a single number (e.g., WWTP influent pathogen concentration). However, such inputs are known to be variable and some are uncertain. The manner in which this variability and uncertainty has been addressed is shown in Table 2-3. The proprietary Excel plug-in product “@RISK” has been used to perform the calculations, incorporating factors that reflect these distributions and inputs (Palisade Corp 2013).<sup>17</sup> The models were run for 1,000 iterations for each virus for each site and for each scenario. On each iteration 100 individuals were ‘exposed’, by taking a random sample from statistical distributions covering the range of possible doses received by individuals ingesting water possibly containing (low levels of) a pathogen.

Note that it can be appropriate to report the results in terms of infection, rather than illness (which is the approach taken for the freshwater component of the New Zealand Guidelines—MfE/MoH 2003). It was also the approach taken in a very recent QMRA study for the Great Lakes (USA—Corsi et al. 2016). These authors opined that “The probability of illness for enteroviruses could not be estimated because illness dose-response and morbidity data were unavailable”. Nevertheless we present an analysis for illness and take the precautionary assumption that all individuals who contract enterovirus infection also become ill. For the other pathogens (adenovirus, Norovirus) we take standard values of the probability of illness, given that infection has occurred. For all pathogens the output metric is therefore an individual’s illness risks, to facilitate comparison with relevant guidelines.<sup>18,19</sup>

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<sup>16</sup> An example is a study by Thebault et al. (2013) of norovirus illness outbreaks among consumers of oysters in southern France.

<sup>17</sup> The @RISK models use named cells as much as possible, to facilitate checking and readability.

<sup>18</sup> There is insufficient time and information to also compute DALY metrics (Disability-Adjusted Life Years) as often used when assessing health risks associated with drinking-water (WHO 2011, chapter 7).

<sup>19</sup> The individual’s illness risk (IIR) is calculated as the total number of predicted illness cases divided by the total number of exposures to potentially contaminated water or shellfish flesh. It represents the risk to an individual swimmer or shellfish consumer on any day, having no prior knowledge of any contamination from the outfall. It is calculated via the Monte Carlo modelling, for which 100 individuals are exposed on each of 1,000 separate days, i.e.,  $10^5$  exposures. The total number of cases is  $1,000m$  where  $m$  is the mean infection case rate over 100 people (readily calculated by the Monte Carlo software—@RISK, Palisade Corp. 2013). So the individual’s infection risk, expressed as a proportion, is  $1,000m/10^5 = m/100$ . When expressed as a percentage,  $IIR = m\%$ .

### 3 Predicted Risk profiles and Individual Illness Risks (IIR)

The following tables report the predicted number of illness cases (out of 100 people exposed on any random occasion) and the IIR.

**Table 3-1: IIR results for adenovirus and enterovirus for seven virus removal orders, assuming typical illness patterns in the Rotorua community.**

Adenovirus ("A") for 7 log <sub>10</sub> removal orders								Enterovirus ("E") for 7 log <sub>10</sub> removal orders							
Statistic	A2_ill	A3_ill	A4_ill	A5_ill	A6_ill	A7_ill	A8_ill	E2_ill	E3_ill	E4_ill	E5_ill	E6_ill	E7_ill	E8_ill	
Min	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
5%ile	1	0	0	0	0	0	0	0	0	0	0	0	0	0	
10%ile	1	0	0	0	0	0	0	1	0	0	0	0	0	0	
15%ile	2	0	0	0	0	0	0	1	0	0	0	0	0	0	
20%ile	2	0	0	0	0	0	0	1	0	0	0	0	0	0	
25%ile	2	0	0	0	0	0	0	2	0	0	0	0	0	0	
30%ile	3	0	0	0	0	0	0	2	0	0	0	0	0	0	
35%ile	3	0	0	0	0	0	0	2	0	0	0	0	0	0	
40%ile	3	0	0	0	0	0	0	2	0	0	0	0	0	0	
45%ile	4	0	0	0	0	0	0	3	0	0	0	0	0	0	
50%ile	4	0	0	0	0	0	0	3	0	0	0	0	0	0	
55%ile	4	0	0	0	0	0	0	3	0	0	0	0	0	0	
60%ile	4	0	0	0	0	0	0	4	0	0	0	0	0	0	
65%ile	5	0	0	0	0	0	0	4	0	0	0	0	0	0	
70%ile	5	1	0	0	0	0	0	4	0	0	0	0	0	0	
75%ile	6	1	0	0	0	0	0	5	1	0	0	0	0	0	
80%ile	6	1	0	0	0	0	0	5	1	0	0	0	0	0	
85%ile	7	1	0	0	0	0	0	5	1	0	0	0	0	0	
90%ile	7	1	0	0	0	0	0	6	1	0	0	0	0	0	
95%ile	9	2	0	0	0	0	0	7	2	0	0	0	0	0	
96%ile	9	2	0	0	0	0	0	8	2	0	0	0	0	0	
97%ile	10	2	1	0	0	0	0	8	2	1	0	0	0	0	
98%ile	10	2	1	0	0	0	0	9	2	1	0	0	0	0	
99%ile	12	3	1	0	0	0	0	10	2	1	0	0	0	0	
99.5%ile	13	3	1	0	0	0	0	11	3	1	0	0	0	0	
99.9%ile	15	3	2	1	0	0	0	13	4	1	1	0	0	0	
Max	20	5	2	1	1	1	0	15	4	3	2	1	0	0	
IIR(%)	4.1873	0.4128	0.0392	0.0038	0.0003	0.0001	0	3.2749	0.35	0.0366	0.0046	0.0006	0	0	

**Table 3-2: IIR results for aggregated and disaggregated norovirus for seven virus removal orders, assuming typical illness patterns in the Rotorua community.**

Norovirus ("N"), disaggregated, for 7 log10 removal orders								Norovirus ("N"), aggregated, for 7 log10 removal orders						
Statistic	Ndis2_ill	Ndis3_ill	Ndis4_ill	Ndis5_ill	Ndis6_ill	Ndis7_ill	Ndis8_ill	Nagg2_ill	Nagg3_ill	Nagg4_ill	Nagg5_ill	Nagg6_ill	Nagg7_ill	Nagg8_ill
Min	2	0	0	0	0	0	0	0	0	0	0	0	0	0
5%ile	15	2	0	0	0	0	0	0	0	0	0	0	0	0
10%ile	17	3	0	0	0	0	0	0	0	0	0	0	0	0
15%ile	19	4	0	0	0	0	0	0	0	0	0	0	0	0
20%ile	20	5	0	0	0	0	0	0	0	0	0	0	0	0
25%ile	21	5	0	0	0	0	0	0	0	0	0	0	0	0
30%ile	22	6	0	0	0	0	0	0	0	0	0	0	0	0
35%ile	22	6	0	0	0	0	0	0	0	0	0	0	0	0
40%ile	23	7	0	0	0	0	0	0	0	0	0	0	0	0
45%ile	24	7	1	0	0	0	0	0	0	0	0	0	0	0
50%ile	24	8	1	0	0	0	0	0	0	0	0	0	0	0
55%ile	25	8	1	0	0	0	0	0	0	0	0	0	0	0
60%ile	26	9	1	0	0	0	0	0	0	0	0	0	0	0
65%ile	26	10	1	0	0	0	0	0	0	0	0	0	0	0
70%ile	27	10	1	0	0	0	0	0	0	0	0	0	0	0
75%ile	28	11	2	0	0	0	0	0	0	0	0	0	0	0
80%ile	29	12	2	0	0	0	0	0	0	0	0	0	0	0
85%ile	30	13	2	0	0	0	0	0	0	0	0	0	0	0
90%ile	31	14	3	0	0	0	0	1	0	0	0	0	0	0
95%ile	33	16	3	1	0	0	0	1	0	0	0	0	0	0
96%ile	33	17	4	1	0	0	0	1	0	0	0	0	0	0
97%ile	34	19	4	1	0	0	0	1	0	0	0	0	0	0
98%ile	35	21	5	1	0	0	0	1	0	0	0	0	0	0
99%ile	36	24	7	1	1	0	0	2	1	0	0	0	0	0
99.5%ile	37	26	8	2	1	0	0	2	1	0	0	0	0	0
99.9%ile	40	29	11	2	1	1	0	3	1	1	0	0	0	0
Max	47	33	14	3	2	1	1	4	1	1	1	0	0	0
IIR(%)	24.161	8.3762	1.0365	0.1052	0.0121	0.0013	0.0003	0.1535	0.0153	0.0014	0.0001	0	0	0



**Table 3-3: IIR results for adenovirus and enterovirus for seven virus removal orders, assuming outbreak illness patterns in the Rotorua community.**

Adenovirus ("A") for 7 log10 removal orders								Enterovirus ("E") for 7 log10 removal orders							
Statistic	A2_ill	A3_ill	A4_ill	A5_ill	A6_ill	A7_ill	A8_ill	E2_ill	E3_ill	E4_ill	E5_ill	E6_ill	E7_ill	E8_ill	
Min	10	0	0	0	0	0	0	6	0	0	0	0	0	0	
5%ile	19	2	0	0	0	0	0	14	1	0	0	0	0	0	
10%ile	23	3	0	0	0	0	0	18	2	0	0	0	0	0	
15%ile	25	3	0	0	0	0	0	21	2	0	0	0	0	0	
20%ile	26	4	0	0	0	0	0	23	3	0	0	0	0	0	
25%ile	27	4	0	0	0	0	0	24	3	0	0	0	0	0	
30%ile	28	5	0	0	0	0	0	26	3	0	0	0	0	0	
35%ile	29	5	0	0	0	0	0	27	4	0	0	0	0	0	
40%ile	30	5	0	0	0	0	0	28	4	0	0	0	0	0	
45%ile	31	6	0	0	0	0	0	29	5	0	0	0	0	0	
50%ile	32	6	0	0	0	0	0	31	5	0	0	0	0	0	
55%ile	33	7	1	0	0	0	0	32	5	0	0	0	0	0	
60%ile	33	7	1	0	0	0	0	33	6	1	0	0	0	0	
65%ile	34	8	1	0	0	0	0	35	6	1	0	0	0	0	
70%ile	35	8	1	0	0	0	0	36	7	1	0	0	0	0	
75%ile	36	9	1	0	0	0	0	37	7	1	0	0	0	0	
80%ile	38	9	1	0	0	0	0	39	8	1	0	0	0	0	
85%ile	39	10	2	0	0	0	0	40	8	2	0	0	0	0	
90%ile	41	12	2	1	0	0	0	43	10	2	1	0	0	0	
95%ile	46	16	4	1	0	0	0	51	16	4	2	0	0	0	
96%ile	47	38	13	1	0	0	0	97	79	26	6	1	0	0	
97%ile	49	42	22	2	0	0	0	99	86	47	10	1	0	0	
98%ile	51	47	28	6	1	0	0	99	93	57	16	2	0	0	
99%ile	53	50	36	8	1	0	0	100	96	69	20	2	0	0	
99.5%ile	55	52	39	10	1	0	0	100	97	73	22	3	1	0	
99.9%ile	57	55	43	12	3	1	0	100	98	77	27	4	1	1	
Max	59	61	44	14	3	1	0	100	99	79	29	5	1	1	
IIR(%)	32.053	8.174	1.824	0.316	0.036	0.002	0	33.154	8.999	2.977	0.726	0.084	0.01	0.002	

**Table 3-4: IIR results for aggregated and disaggregated norovirus for seven virus removal orders, assuming outbreak illness patterns in the Rotorua community.**

Norovirus ("N"), disaggregated, for 7 log10 removal orders								Norovirus ("N"), aggregated, for 7 log10 removal orders							
Statistic	Ndis2_ill	Ndis3_ill	Ndis4_ill	Ndis5_ill	Ndis6_ill	Ndis7_ill	Ndis8_ill	Nagg2_ill	Nagg3_ill	Nagg4_ill	Nagg5_ill	Nagg6_ill	Nagg7_ill	Nagg8_ill	
Min	15	4	0	0	0	0	0	0	0	0	0	0	0	0	
5%ile	22	15	2	0	0	0	0	0	0	0	0	0	0	0	
10%ile	24	17	3	0	0	0	0	0	0	0	0	0	0	0	
15%ile	25	19	4	0	0	0	0	0	0	0	0	0	0	0	
20%ile	26	20	4	0	0	0	0	0	0	0	0	0	0	0	
25%ile	27	21	5	0	0	0	0	0	0	0	0	0	0	0	
30%ile	27	22	6	0	0	0	0	0	0	0	0	0	0	0	
35%ile	28	22	6	0	0	0	0	1	0	0	0	0	0	0	
40%ile	29	23	7	0	0	0	0	1	0	0	0	0	0	0	
45%ile	29	24	7	1	0	0	0	1	0	0	0	0	0	0	
50%ile	30	24	8	1	0	0	0	1	0	0	0	0	0	0	
55%ile	30	25	8	1	0	0	0	1	0	0	0	0	0	0	
60%ile	31	26	9	1	0	0	0	1	0	0	0	0	0	0	
65%ile	31	26	9	1	0	0	0	2	0	0	0	0	0	0	
70%ile	32	27	10	1	0	0	0	2	0	0	0	0	0	0	
75%ile	33	28	11	2	0	0	0	2	0	0	0	0	0	0	
80%ile	34	28	11	2	0	0	0	2	0	0	0	0	0	0	
85%ile	35	29	13	2	0	0	0	3	0	0	0	0	0	0	
90%ile	36	31	14	2	0	0	0	3	1	0	0	0	0	0	
95%ile	38	33	16	3	1	0	0	4	1	0	0	0	0	0	
96%ile	38	33	17	3	1	0	0	4	1	0	0	0	0	0	
97%ile	39	34	19	4	1	0	0	5	1	0	0	0	0	0	
98%ile	40	35	21	5	1	0	0	6	1	0	0	0	0	0	
99%ile	40	36	23	6	1	0	0	7	2	1	0	0	0	0	
99.5%ile	41	37	25	7	1	1	0	9	2	1	0	0	0	0	
99.9%ile	43	40	27	9	2	1	0	10	3	1	0	0	0	0	
Max	49	40	28	9	2	2	0	12	3	2	0	0	0	0	
IIR(%)	29.666	24.051	8.234	1.009	0.095	0.011	0	1.467	0.148	0.014	0	0	0	0	

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## 5 Acknowledgements

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## 6 Glossary of abbreviations and terms

Aetiological agent	Microorganisms and microbial toxins that cause disease in humans.
Beta-Binomial dose-response curve	A mathematically-derived infection dose-response curve for variable infectivity, in which individual doses are known.
Beta-Poisson dose-response curve	A mathematically-derived infection dose-response curve for variable infectivity, in which only mean doses are known.
Conditional illness probability	The probability of illness at a given dose given that infection has already occurred.
Conditional infection dose-response models	The (simpler) mathematical form of a dose-response equation that results when individual doses are known. (More complicated mathematical functions arise when individual doses are not known).
Hypergeometric functions	Mathematical equations that defy simple calculation, yet are important in the analysis of clinical trial data and outbreak data for the infection response of a population exposed to a pathogen, and where individual doses are randomly distributed about a known mean value.
Illness ID <sub>50</sub>	The dose required to cause illness in 50% of an exposed population, who are already infected.
Infection ID <sub>50</sub>	The dose required to cause infection in 50% of an exposed population.
PCR	Polymerase Chain Reaction, a molecular technique for virus enumeration using DNA segment matching.
QMRA	Quantitative Microbial Risk Assessment.
RT-qPCR	Reverse-transcription quantitative PCR, used for RNA viruses.
Sequelae	An illness that is the result of a previous disease.
Simple binomial dose-response curve	A mathematically-derived infection dose-response curve for constant infectivity, in which individual doses are known.
Simple exponential dose-response curve	A mathematically-derived infection dose-response curve for constant infectivity, in which only mean doses are known.
TCID <sub>50</sub>	Median Tissue Culture Infectious Dose: A laboratory culture technique measuring the amount of virus that produces a cytopathic effect in 50% of cell cultures inoculated.
Virion	Shorthand for “virus particle”.

## Appendix A Virus characteristics

### Adenoviruses

Respiratory viruses, particularly some adenoviruses, may also need to be considered within a QMRA. Respiratory symptoms (via inhalation of contaminated water when water skiing, or inhaling surf-generated aerosols) are sometimes associated with contact with wastewater-impacted coastal waters (WHO, 2003). In particular, a New Zealand epidemiological study at seven coastal beaches found a respiratory effect associated with the faecal indicator bacterium enterococci (McBride et al. 1998). Respiratory-associated viruses are probably the commonest causes of acute respiratory infections, reportedly causing around 70% of acute sore throats (Mims et al. 2004). They can be particularly resistant to disinfection (Gerba et al. 2003, Thompson et al. 2003). However, while adenoviruses are commonly found in water (Horwitz 2001), including wastewater, many strains give rise to gastrointestinal illness (e.g., the 40/41 strain complex), with a rather smaller proportion associated with respiratory symptoms. So we should note that we have clinical trial information available only for the respiratory-illness-causing adenovirus 4 (Couch et al. 1966a&b, 1969) for which a dose-response model has been developed (Haas et al. 1999). We can expect that people are more vulnerable to respiratory agents than to gastrointestinal agents, because the human body's defences to the latter are more formidable. Fong et al. (2010) found only 3% of wastewater adenoviruses were type 4. So QMRA studies that apply the adenovirus 4 infection dose-response model to all adenoviruses (Gerba et al. 1996, Crabtree et al. 1997) have over-estimated health risk.

Other QMRA studies in New Zealand have predicted illness via ingestion among recreational water users near marine outfalls to be rather higher than illness-via-inhalation (Stott & McBride 2011). A recent study of wet weather bypass flows at Moa Point, Wellington, has included consideration of respiratory effects, using Fong's results (Crawford et al. 2014).

### Enteroviruses

Enterovirus (EV) is a single-stranded member of the picornavirus family, containing over 70 serotypes.<sup>21</sup> Although it was originally classified into 4 groups, polioviruses, coxsackie A viruses, coxsackie B viruses, and echoviruses, molecular characterisation has led to their reclassification into an enterovirus genus that includes 12 species: enterovirus A-H, J and Rhinovirus A-C. Human species of enterovirus are grouped into the four EV species A-D and the three Rhinovirus groups A-C.

Enteroviruses are often found in respiratory secretions (e.g., saliva, nasal mucus) and stools of infected persons. Poliovirus, coxsackie and echovirus can be spread through the faecal-oral route. Infection can result in a wide variety of symptoms ranging from mild respiratory illness (common cold), hand, foot and mouth disease, acute haemorrhagic conjunctivitis, aseptic meningitis, myocarditis, severe neonatal sepsis-like disease, and acute flaccid paralysis. Enteroviruses are distributed worldwide and are influenced by season and climate. Infections can show a seasonal pattern, with enterovirus prevalence peaking in summer and early fall in temperate areas, while no discernible seasonal trend is evident in tropical and semitropical areas.

A comparison with literature data found that E-30 (echovirus 30) was the most prevalent type detected internationally (Janes et al. 2014). Generally, enterovirus B viruses (in particular echoviruses) were the most frequently detected. Age distribution patterns were observed with 30–74% of all isolates detected in young children (< 5 years).

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<sup>21</sup> <http://www.picornastudygroup.com/types/enterovirus/enterovirus.htm>

Surveillance and monitoring of enteroviruses has traditionally been based on culturing and serotyping. However, it is likely that concentrations may be under-reported due to differences in cell culture sensitivities (see Schiff et al. 1984a&b). Current advances in molecular techniques using RT-PCR for detection followed by sequencing of the capsid genes for typing is now the method typically used (Benschop et al. 2010).

## Noroviruses

Noroviruses are a principal cause of viral gastroenteritis. They are single-stranded RNA viruses that have been classified into 5 genogroups (GI to GV). Strains I, II and IV can infect humans (particularly strain GII, see Matthews et al. 2012), while GIII infects bovine species and GV has recently been identified in mice. The GI viruses are highly infectious for a proportion of the population (Teunis et al. 2008) and spread easily by direct person-to-person or person-surface-person contact. By analogy, the GII genogroup exhibits the same behaviour. They also can be associated with waterborne gastroenteritis (Parshionikar et al. 2003) or shellfish-associated gastroenteritis (Lees et al. 1995, Thebault et al. 2013)<sup>22</sup> and are therefore a hazard to recreational water users (Gray et al. 1997). They have been detected in both raw and treated wastewaters (Nordgren et al. 2009), with strains of GI and GII predominating in human-derived wastewater that are typically very similar to human strains circulating in the population (van den Berg et al. 2005). Therefore, the public may be at appreciable risk whenever there is exposure to human wastes (animal viruses are generally thought to be not infectious to humans, and so other animal pathogens—bacteria and protozoa—come into play). For the purposes of the QMRA, noroviruses therefore represent the primary potential risk of infection from human-derived wastewaters via ingestion for primary contact users, such as swimmers, surfers and body boarders.

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<sup>22</sup> These authors considered both infection and illness.



## Appendix B Dose-response functions

### For infection

Standard clinical trial procedures involve challenging groups of volunteers with aliquots taken from serially-diluted preparations whose well-mixed concentrations are measured. Doses in individuals' challenges are not measured. Consequently only the average dose given to each member of a group is known. Nevertheless, by making two simple assumptions the mathematical form of the infection dose-response equation can be obtained (Haas et al. 1999, McBride 2005a):

1. The "single-hit" hypothesis: That a single pathogen, surviving the body's barriers (e.g., acidic digestion system) and reaching a potential infection site, is sufficient to cause infection.
2. Poisson distribution of pathogens in the preparation—as is appropriate for a random well-mixed population.

The mathematical result, after averaging across each group's individual Poisson-distributed doses, is the single-parameter "simple exponential" equation

$$\text{Pr}_{\text{inf}}(d) = 1 - e^{-rd} \quad (1)$$

where  $d$  is the average doses given to each group, "e" is the standard exponential number (the base of natural logarithms,  $e = 2.7183\dots$ ), and  $r$  is the probability that a pathogen survives the body's defences and reaches an infection site.

Sometimes host-pathogen interactions are such that a constant value of  $r$  is implausible (e.g., because of differential immunity, or varying pathogen virulence, as indicated by lack of fit to the single-parameter model). In that case  $r$  is replaced by a standard two-parameter beta distribution with shape parameter  $\alpha$  and location parameter  $\beta$ . The mathematical result is the much-more-difficult-to-evaluate<sup>23</sup> Kummer hypergeometric function (denoted as  ${}_1F_1$ ):

$$\text{Pr}_{\text{inf}}(d) = 1 - {}_1F_1(\alpha, \alpha + \beta, -d) \quad (2)$$

For obvious reasons this can be called the "beta-Poisson" equation.<sup>24</sup> Fortunately in many cases we find that  $\beta \gg 1$  and  $\alpha \ll \beta$ , in which case this equation can be well-approximated by the following equation (confusingly, also called "beta-Poisson")

$$\text{Pr}_{\text{inf}} = 1 - \left(1 + \frac{d}{\beta}\right)^{-\alpha} \quad (3)$$

However this approximation is inadequate for noroviruses because the fitted parameter doublet ( $\alpha = 0.04$  and  $\beta = 0.055$ , Teunis et al. 2008) constitute a serious breach of the approximation-validity criteria ( $\alpha \ll \beta$ ,  $\beta \gg 1$ ). Analysis of clinical trial data for noroviruses therefore calls for specialist software that can evaluate (2), as reported by Teunis et al. 2008, Thebault et al. (2013).

<sup>23</sup> Equation (2) can't be evaluated in Excel.

<sup>24</sup> Because a two-parameter ( $\alpha$  and  $\beta$ ) beta distribution is used instead of the single parameter  $r$  and the doses are assumed random, i.e., Poisson-distributed. Strictly,  $\beta$  is not properly a location parameter for equation (2), but it is for its approximation equation (3) (because  $d$  is simply divided by  $\beta$  in that equation: increasing the value of  $\beta$  shifts the curve to the right).

### Simplifying the infection dose-response calculations for QMRA

Good QMRA practice, especially for virulent pathogens, is to "expose" *multiple* people on each exposure occasion.<sup>25</sup> In that case the individual doses are known (i.e., are calculated and assigned to individuals by the model) so that there is no need for Poisson-averaging. This somewhat simplifies the mathematical development of the infection dose-response formulae such that for constant  $r$  the simple one-parameter exponential model is replaced by the simple binomial model

$$\Pr_{\text{inf}} = 1 - (1 - r)^i \quad (4)$$

where  $i$  is the individual's dose.

Also, the two-parameter beta-Poisson model (the  ${}_1F_1$  functional form) is replaced by the "beta-binomial" model

$$\Pr_{\text{inf}} = 1 - \frac{B(\alpha, \beta + i)}{B(\alpha, \beta)} \quad (5)$$

where  $B$  is the standard beta function (Abramowitz & Stegun 1972) and  $\alpha$  and  $\beta$  are as defined previously. This equation can be simply evaluated in Excel.<sup>26</sup>

These two equations have been described by Haas (2002) as conditional infection dose-response models, the condition being that individual doses are known.

The following figures (Figure B-1a&b) give examples of these functions for adenovirus 4 and for Norwalk virus, for both conditional and unconditional infection dose-response models.

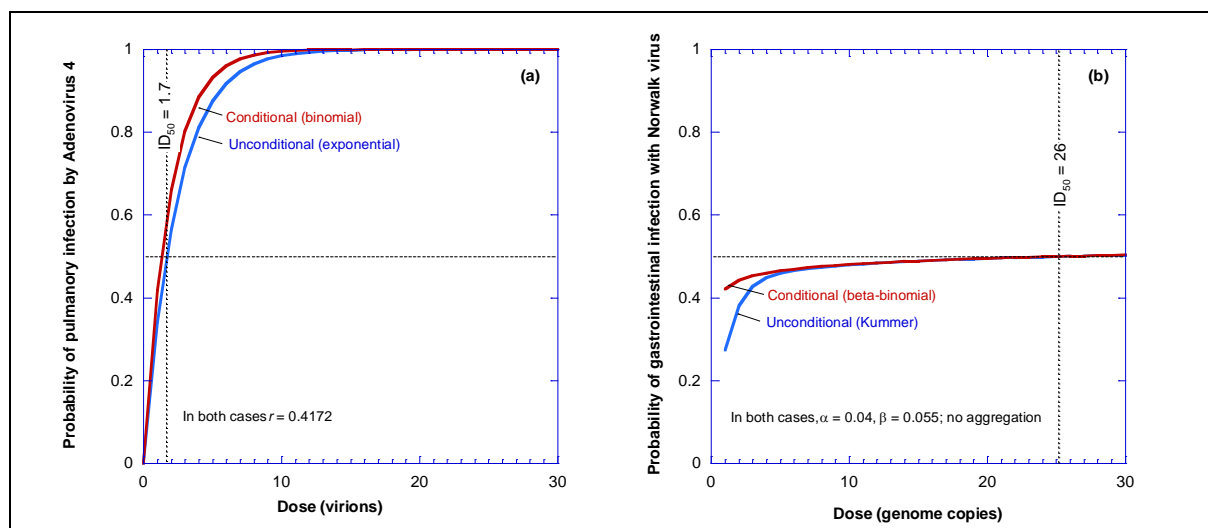


Figure B-1: Conditional and unconditional infection dose-response curves for: (a) single-parameter models for adenovirus 4, and (b) double-parameter models for Norwalk virus (only for susceptible individuals).

<sup>25</sup> To not do so gives rise to implausible risk profiles. For example if only one individual is exposed per exposure occasion—as a representative of a group visiting a contaminated beach—and if the probability of infection given ingestion of one pathogen is high (say, 20%), then probabilities of infection between 0% and 20% are impossible. The resulting risk profile becomes extremely jagged (McBride 2005b). In such cases exposing a group of people per exposure occasion (say, 100), each with different doses (some swim for a few minutes, others for an hour or so), allows many values between 0 and 20% to be calculated.

<sup>26</sup> To do so we note that  $B(\alpha, \beta) = \Gamma(\alpha)\Gamma(\beta)/\Gamma(\alpha+\beta)$ , where  $\Gamma$  is the standard Gamma function (Abramowitz & Stegun 1972). Standard Excel includes the natural logarithm of the gamma function (as the function 'GAMMALN'), so that we can derive:  $\Pr = 1 - \text{EXP}\{\text{GAMMALN}(\beta+i) + \text{GAMMALN}(\alpha+\beta) - [\text{GAMMALN}(\alpha+\beta+i) + \text{GAMMALN}(\beta)]\}$ .

These graphs highlight some important features of infection dose-response curves:

- The single-parameter models (e.g., Figure B-1a) rise inexorably to unit probability, precisely because their common parameter ( $r$ ) is constant.
- The double-parameter models (e.g., Figure B-1b) “flatten out” well before reaching unit probability.<sup>27</sup>
- Whilst the relatively high infection ID<sub>50</sub> for Norwalk virus (26 genome copies among susceptible individuals) occurs on the flattened top of its dose-response curve, infection probabilities are still appreciable at much lower doses.<sup>28</sup>
- The unconditional curves have a jagged profile around the conditional forms, yet deploying the latter in a QMRA gives rise to the same averaged risk.<sup>29</sup>
- Whilst the adenovirus 4 infection dose-response curve is in all respects more severe than that for Norwalk virus, for two reasons that doesn’t mean that it is the most severe pathogen:
  - i. adenoviruses that can cause respiratory ailments are a minor part of the total adenovirus population in sewage,<sup>30</sup> with most causing gastro-intestinal illness
  - ii. exposure to respiratory adenoviruses (via inhalation, e.g., whilst surfing) tends to be lower than ingestion of water whilst swimming.<sup>31</sup>

However, having double-stranded DNA, adenoviruses are more resistant to disinfection processes.

## For illness

Some individuals who become infected (e.g., as measured by serological response, or by evidence of pathogen shedding) may not go on to exhibit symptoms, i.e., they are asymptomatic. In that case, to obtain the unconditional probability of illness (given dose) we first need to calculate the conditional probability of illness given infection for each dose, denoted as  $Pr_{ill|inf}$ . The probability of illness is calculated as:

$$Pr_{ill} = Pr_{ill|inf} Pr_{inf} \quad (6)$$

Two common approaches are used for the conditional illness function:

### Hazards model

Teunis et al. (1999) developed hazard models for the illness given infection, with two forms

<sup>27</sup> In fact these models approach unit probability only for enormous doses.

<sup>28</sup> The “flat top” is caused by the variable host-pathogen interactions, including a proportion of exposed population who high (but incomplete) immune. There is also another group who are completely immune.

<sup>29</sup> That’s because applying the unconditional form to a single individual representing a group of people, as is common practice, doesn’t capture the fact that, by good luck, some people at a beach will avoid exposure whilst the averaged dose is above zero (McBride 2005b).

<sup>30</sup> Typically respiratory serotypes are detected less frequently than adenovirus F serotypes and so the gastro-intestinal (GI) disease-causing serotypes tend to predominate in sewage studies (Osuolale & Okoh 2015). However, a proportion of respiratory versus GI serotypes detected will depend on the cell line used for culture assays and the target primers for molecular methods. For example, Hewitt et al. (2011) used cell line 594 and reported that culturable adenoviruses were mainly A-E types (which are respiratory and conjunctivitis serotypes) and there was still around 3 log presence in effluents.

<sup>31</sup> Water-contact-related respiratory illness is an area worthy of further research, particularly in the light of the respiratory illness rates reported in the one New Zealand epidemiological study on this matter—McBride et al. (1998). In that study (at seven New Zealand beaches) those rates were generally more prominent than gastrointestinal rates, a phenomenon that is not fully understood.

$$\text{Decreasing hazard} \quad \Pr_{\text{illnf}}(d) = 1 - \left(1 + \frac{\eta}{d}\right)^{-r} \quad (7)$$

and

$$\text{Increasing hazard} \quad \Pr_{\text{illnf}}(d) = 1 - (1 + \eta d)^{-r} \quad (8)$$

where  $\eta$  is a location parameter, and  $r$  is a shape parameter.<sup>32</sup>

#### *Dose independence*

Existing models of the conditional probabilities of illness (the condition being that infection has already occurred) are held in some doubt internationally. For example, the norovirus model (Teunis et al. 2008) predicts substantial infection probabilities at very low doses, but predicts substantial illness probabilities (among the infected) only at very high doses. A large body of work has taken the view that the conditional probability of illness-given-infection should be independent of dose—Schoen & Ashbolt (2010), Soller et al. (2010, 2015), Viau et al. (2011) and Boehm et al. (2015). Indeed, that approach is endorsed by WHO (2011), with the result that for the pathogens considered here the conditional illness probabilities are on the order of ½.

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<sup>32</sup> The decreasing hazards model has only been reported for a clinical trial on adults exposed to *Campylobacter* (Teunis et al. 1999): All other conditional illness models that I am aware of infer an increasing hazards model, including a *Campylobacter* outbreak study for children (Teunis et al. 2005).

## Appendix C Echovirus 12 clinical trial data analysis

Echovirus is a member of the enterovirus family. Haas et al. (1999) reported fitting a one-parameter simple exponential model to clinical trial data for an echovirus 12 study (Akin 1981),<sup>33</sup> with an estimated infection  $ID_{50} = 54$  virions, corresponding to their calibrated  $r$  value of 0.0128.<sup>34</sup> Haas (1983) had earlier fitted a slightly different value to the Akin data, with  $r = 0.012$  (giving infection  $ID_{50} = 58$ ) and also a two-parameter beta-Poisson curve (with  $\alpha = 1.3$  and  $\beta = 75$ ), so that the infection  $ID_{50} [= \beta(2^{1/\alpha} - 1)] = 53$ . Clearly, these approaches give consistent results with an infection  $ID_{50}$  about 50.

The beta-Poisson result was used in the QMRA performed for the Mangere wastewater treatment upgrade (DRG 2002, Simpson et al. 2003), this choice being particularly influenced by the observation that enterovirus illness can give rise to more serious consequences (i.e., sequelae) relative to other virus groups.

Akin's data were in fact preliminary results from an ongoing clinical trial, full results of which were reported three years later in Schiff et al. (1984a&b). Their 1984a paper is the proceedings of a conference held two years earlier in Herzliya Israel. It contains the Akin data. But the 1984b document (a peer-reviewed journal paper) multiplied all the doses, including those reported by Akin, by a factor of 33, to account for the re-analysis of the stock dose suspension using a more sensitive cell line<sup>35</sup>. These published data were analysed by Teunis et al. (1996) giving rise to a two-parameter "beta-Poisson" model ( $\alpha = 0.401$ ,  $\beta = 227.2$ , as reported by Teunis et al. 1996) and a higher infection  $ID_{50} = 1052$  virions.<sup>36</sup>

We propose to use the beta-Poisson model ( $\alpha = 1.3$  and  $\beta = 75$ , with infection  $ID_{50} = 53$  virions). Note that this conflicts with the approach taken in the increasingly-influential CAMRA website<sup>37</sup> ( $\alpha = 1.06$  and  $\beta = 171.3$ ), giving rise to an infection  $ID_{50} = 922$ . This has implications for the enterovirus concentrations to be presented to this dose-response function in the QMRA calculations.<sup>38</sup>

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<sup>33</sup> This widely-quoted paper (Akin 1981) seems to have been read by only a few, given its appearance only in the "grey literature", decades past. The author of this report has a copy, courtesy of Professor Haas (Drexel University), which is available on request.

<sup>34</sup> For the simple exponential model, algebraic manipulation shows that  $ID_{50} = -\ln(1/2)/r \approx 0.693/r$ .

<sup>35</sup> At page 864 of Schiff et al. (1984b): "The original plaque assay used for determination of the titer of the echovirus-12 pool and of the various challenge doses administered to volunteers was based on the use of LLC-MK<sub>2</sub> cells and an agar overlay procedure; in the present study this assay was shown to be significantly less sensitive than the plaque neutralization assay involving RD cells and a soft agar overlay procedure. The latter system increased the plaquing efficiency of the challenge virus by 33-fold."

<sup>36</sup> For the approximate beta-Poisson model, algebraic manipulation shows that  $ID_{50} = \beta(2^{1/\alpha} - 1)$ .

<sup>37</sup> Center for Advancing Microbial Risk Assessment Not [http://qmrwiki.canr.msu.edu/index.php/Dose\\_Response](http://qmrwiki.canr.msu.edu/index.php/Dose_Response)

<sup>38</sup> The adopted dose-response function refers to echovirus 12 data gathered using the "LLC-MK<sub>2</sub>" cell line (Schiff et al. 1984a). The CAMRA dose-response function refers to data re-analysed using "RD" cell line. Comparison of dose-response functions for other members of the enterovirus group (e.g., polio virus, hepatitis A, coxsackie) indicates that  $ID_{50}$  of the order of 50 is more tenable than of the order of 1000.

## Appendix D      Debate about norovirus infection dose-response

We have taken a form of norovirus infection dose-response that has become an “industry standard” in the last five years. It is based on a clinical trial, and is broadly supported by an outbreak study on French oysters (Thebault et al. 2013). That choice reflects a reasonable precautionary stance. Two recent contributors to the journal *Risk Analysis* have presented findings that norovirus may be even more infectious (Messner et al. 2014), or less infectious (Schmidt 2014) than the industry standard dose response, depending largely on the assumed degree of virus aggregation. There is currently much debate about all that. For example, another writer used data from a new clinical trial to claim that norovirus is much less infectious than the industry standard (Atmar et al. 2011, 2014) (this analysis appears to be flawed, as it ignored the role of aggregation, see McBride 2014a).

The role of noroviruses in QMRA will continue to be contentious, not least because a recently published procedure for their enumeration by culture (Jones et al. 2014) supplanted an earlier unsuccessful claim to such a procedure (Straub et al. 2007). This reflects the fact the QMRA is still an emerging discipline, with a number of issues that will take years to resolve. Nonetheless, experience indicates that QMRA is a more informative approach to human health risk assessment relative to that provided by levels of indicator bacteria derived from epidemiological studies at sites generally far-removed from the effects of discharges from large wastewater treatment plants.

## Appendix B    Analysis of Quantitative Microbiological Risk Assessment Inputs and Results



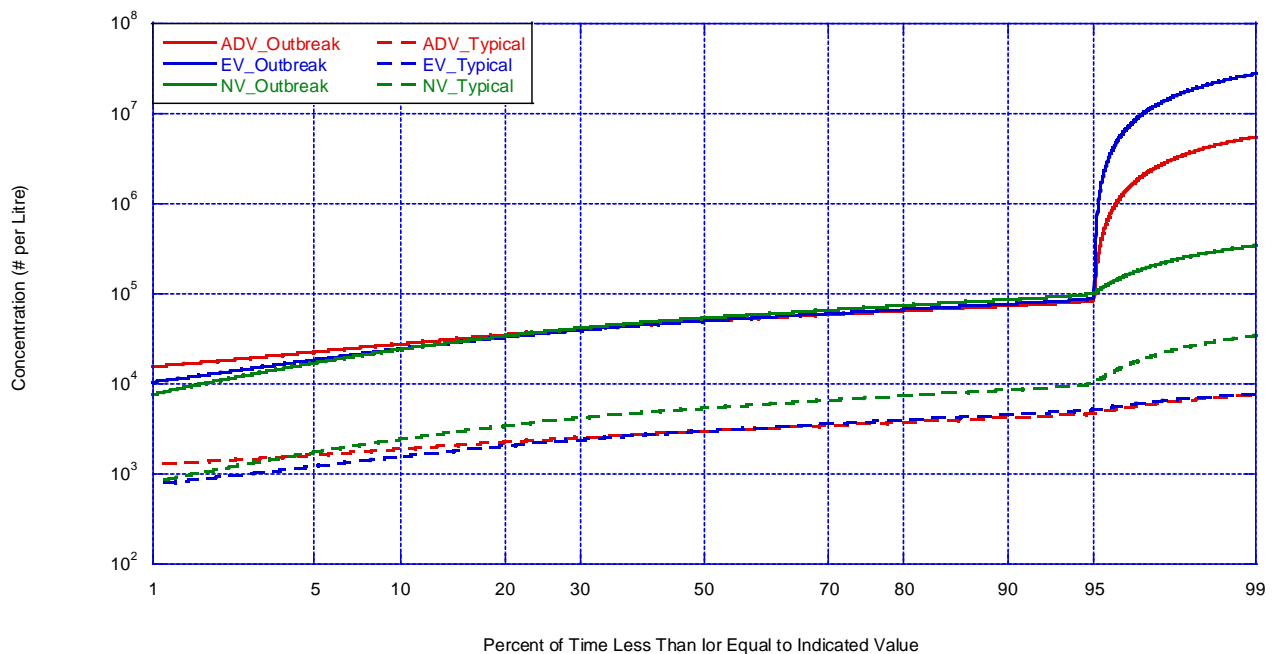


Figure B-1: Distribution over time of the Typical (dotted line) and Outbreak (solid line) Influent Virus Concentrations of Adenovirus, ADV, Enterovirus, EV and Norovirus, NV used in this QMRA.

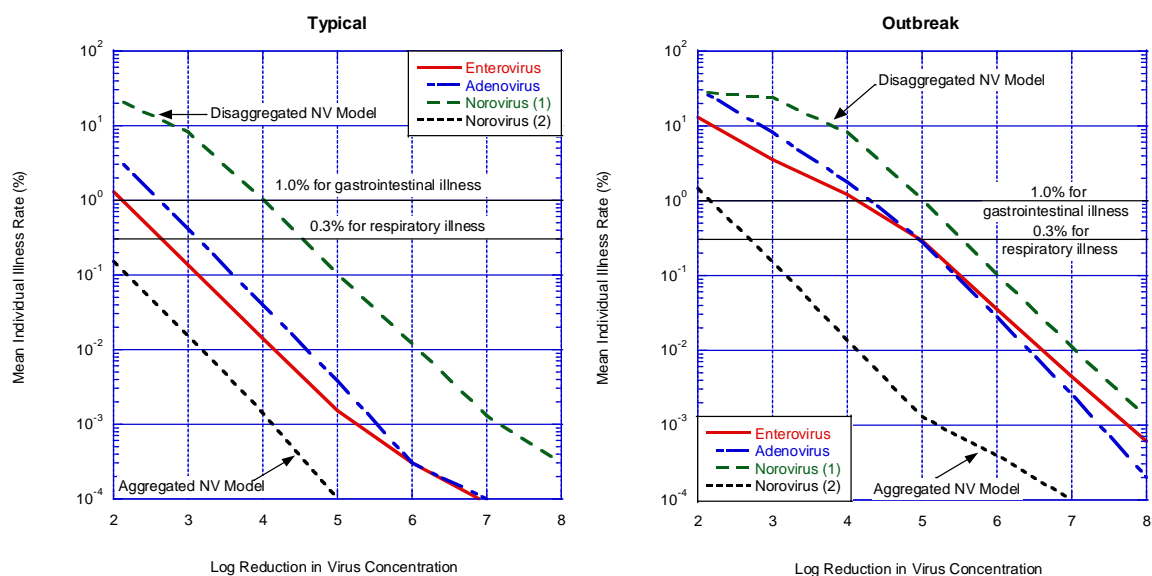


Figure B-2: Individual's Illness Rate (%) Associated with Primary Contact Recreation (Swimming) based on Range of Virus Log Reduction Values for Typical (upper graph) and Outbreak (lower graph) Influent Virus Concentrations of Adenovirus, Enterovirus and Norovirus (aggregated and disaggregated).

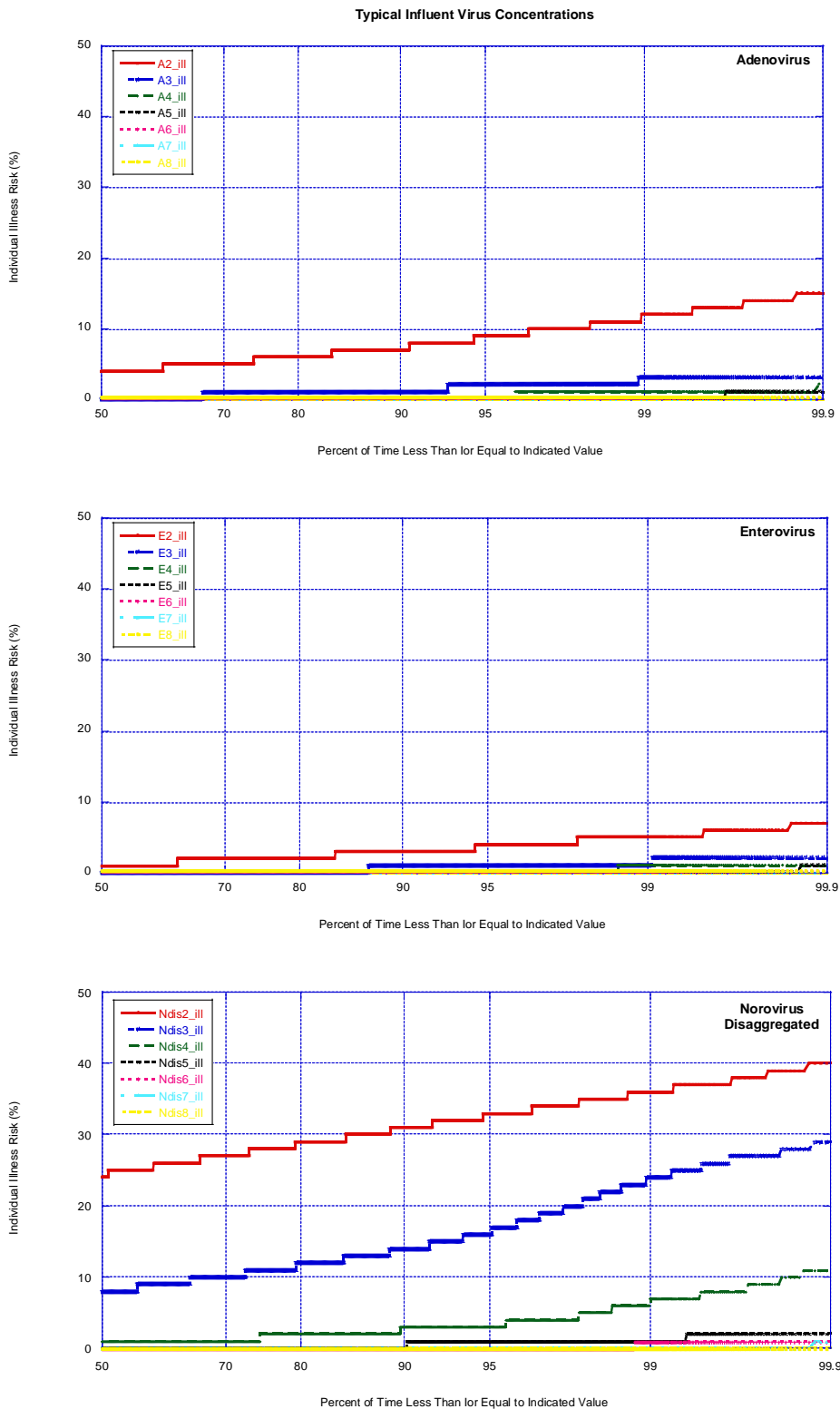


Figure B-3: Distribution over time of the Individual's Illness Rate (%) Associated with Primary Contact Recreation (Swimming) based on Range of Virus Log Reduction Values for Typical Influent Virus Concentrations of Adenovirus, A (upper graph), Enterovirus, E (middle graph), Norovirus disaggregated, Ndis (lower graph).

Note: Virus Log Reduction shown in legend, eg A2\_ill refers to 2 log reduction of adenovirus.

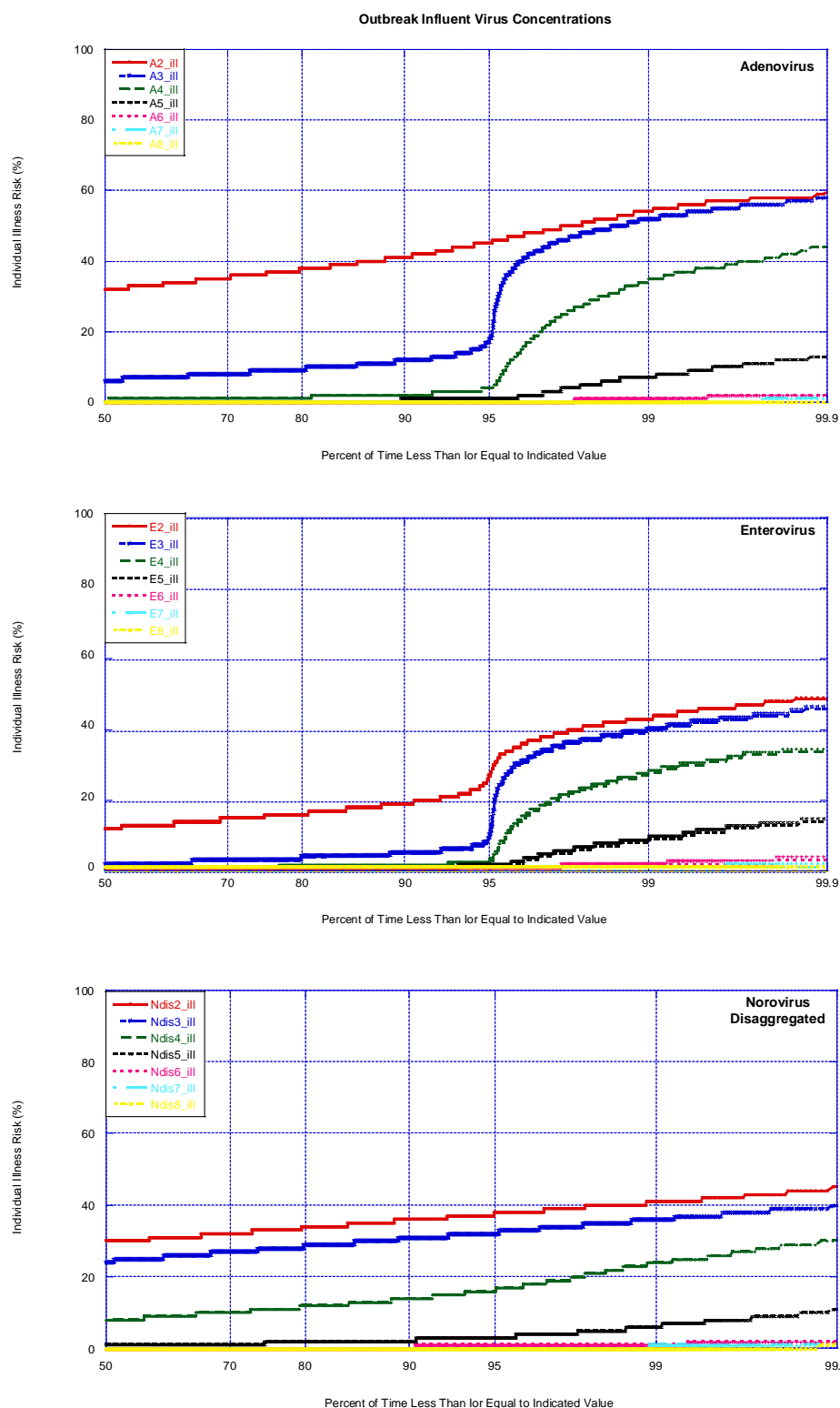


Figure B-4: Distribution over time of the Individual's Illness Rate (%) Associated with Primary Contact Recreation (Swimming) based on Range of Virus Log Reduction Values for Outbreak Influent Virus Concentrations of Adenovirus, A (upper graph), Enterovirus, E (middle graph), Norovirus disaggregated, Ndis (lower graph).

Note: Virus Log Reduction shown in legend, eg A2\_ill refers to 2 log reduction of adenovirus.

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