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Comparing Dose-Response for Infection and Illness in Human Challenge Studies of Norwalk Virus

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Comparing Dose-Response for Infection and Illness in Human Challenge Studies of Norwalk Virus

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2015

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An abstract of
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Abstract

Comparing Dose-Response for Infection and Illness in Human Challenge Studies of Norwalk Virus

By Allison Foster

Noroviruses are the leading cause of acute viral gastroenteritis among all age groups worldwide. This study used both older and recent Norwalk virus (GI.1 norovirus genotype) to model the dose-response relationship for Norwalk virus infection and illness. By comparing data from all published Norwalk virus human challenge studies (N=17) to studies published in 2008 or later (N=5), we assessed whether the 8fIIa dose-response relation has shifted over time. Data from the 17 published challenge studies were used to establish a Beta Poisson dose-response model that accounts for variation in susceptibility among hosts while also accounting for the different inocula used in these studies (8fIIa, 8fIIb, or pool lot number 42399). We found no evidence to suggest a shift in the dose-response model among recent studies compared to all studies and concluded that the concentration of infectious virus in the 8fIIa inoculum has apparently persisted over time. We estimated that the average probability of infection for a single Norwalk virus particle is approximately 0.04, and our estimate of the ID_{50} of Norwalk virus is 5.65×10^3 GEC (95% CI: 687-34,231) among susceptible hosts. These results are consistent with other published estimates of Norwalk virus infectivity and pathogenicity. Future research should focus on expanding existing norovirus dose-response models to incorporate data from human challenge studies using other norovirus genotypes and outbreak data.

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Chapter 1: Literature Review

A. Background

Epidemiology

Noroviruses are the leading cause of acute gastroenteritis and foodborne disease outbreaks in the United States, accounting for more than half of all outbreaks of gastroenteritis each year [1,2]. A study by Hall et al. indicates that the overall disease burden is substantial, estimating that noroviruses cause nearly 19 to 21 million cases of acute gastroenteritis in the U.S. annually, as well as contribute to as many as 71,000 hospitalizations and 800 deaths [3]. Although substantial progress has been made to control diarrheal diseases globally, approximately 18% of diarrheal diseases worldwide can be attributed to norovirus, with high-, middle-, and low-income settings all experiencing similar disease incidences [4]. In developing countries where diarrheal diseases are still a leading cause of death in certain age groups, noroviruses may cause as many as 200,000 deaths in children 5 and younger each year [5].

Previously known as Norwalk or Norwalk-like viruses, noroviruses were first identified in 1968 from an outbreak of gastroenteritis in a school in Norwalk, Ohio. The Norwalk virus was the first prototype norovirus to be identified and described. No etiologic agent could be determined from the 1968 outbreak until Albert Kapikian and his team first observed the 27-nm Norwalk virus particle by examining stool filtrates derived from the outbreak using immune electron microscopy in 1972 [6]. Noroviruses have since proven to be highly contagious, causing numerous outbreaks in a variety of settings worldwide, including hospitals, nursing homes, and cruise ships [7-9]. Once described as

"the perfect human pathogen," increasing understanding of norovirus is of public health concern in order to prevent and control the spread of disease [10].

Symptoms, infection, and illness

Noroviruses infect people of all ages, and symptoms are similar to those of other enteric diseases [11,12]. Clinical norovirus infection is characterized by acute onset of non-bloody diarrhea, vomiting, abdominal cramps, nausea, and low-grade fever. Among primary cases in the 1968 outbreak in Norwalk, the most common symptoms were nausea (98%), vomiting (92%), and abdominal cramps (59%), but 38% of cases also reported diarrhea [13]. Among symptomatic individuals, the incubation period is brief. A systematic review of genotype I and II infections determined that the median incubation period is approximately 1.2 days (95% CI: 1.1 to 1.2 days) [14]. The disease is usually self-limited and typically lasts 12 to 72 hours. One study examining norovirus outbreaks in England found that staff members who were infected experienced a shorter duration of illness compared to patients (2 days compared to 3 days) and also recovered more quickly (3 days compared to 5 days) [15]. Children have also been shown to have longer illness and experience viral shedding longer compared to adults, especially children with immunocompromising conditions [16]. For example, a study among pediatric oncology patients revealed that viral shedding lasted for 23 days on average, while some patients exhibited shedding for 140 days after initial infection [17]. Illness is usually mild for otherwise healthy adults, but more severe illness and death can occur. Some populations may be at greater risk of experiencing additional complications like dehydration and hospitalization, particularly older adults, young children, and individuals with other underlying medical conditions [15,18].

Following infection, an individual may become ill and develop symptoms of acute gastroenteritis; however, human challenge studies indicate that approximately 30% of infected individuals are asymptomatic [19,20]. Despite having no symptoms, these individuals could still be shedding virus particles in stool, meaning they can still transmit the virus. Studies conducted among asymptomatic food handlers in South Korea demonstrated that 1.0 to 3.4% of subjects shed norovirus in stool, while studies in pediatric populations suggest that this number could be as high as 49.2% [21-23]. This poses an additional challenge to controlling and preventing the spread of norovirus in outbreak settings. Ultimately, the overall public health significance of prolonged shedding of symptomatic and asymptomatic individuals is unclear.

Although exposure to a pathogen is necessary for infection to occur, human challenge studies demonstrate that not everyone who is exposed will become infected or develop symptoms of gastroenteritis, even when exposed to high doses [24,25]. Infection with norovirus may offer some short-term protection against future infection for up to 14 weeks, but long-term immunity has not been shown [26]. Certain host characteristics make some individuals more susceptible to norovirus infection. Research suggests that host susceptibility is determined in part by human ABH histo-blood group antigens (HBGAs) because viral infection requires that norovirus particles bind to these attachment receptors [27]. A study conducted in 2002 found that individuals with an O phenotype are more likely to be infected with Norwalk virus compared to other histo-blood groups [28]. More recent studies have shown that some individuals, called secretors (Se+), have a functional fucosyltransferase (FUT2) enzyme that allows virus-like particles (VLPs) to attach to surface epithelial cells of the gastroduodenal junction

and cause either symptomatic or asymptomatic infection, while non-secretor (Se-) individuals did not. Challenge studies suggest that secretor status is an important predictor of susceptibility among Norwalk and GII.4 genotypes, but no association has been found for the Snow Mountain virus genotype [25,29,30]. However, challenge studies of Norwalk and GII.4 genotypes demonstrate that some Se+ individuals were still resistant to infection even at moderate doses, suggesting that additional factors are likely important in understanding individual risk and merit further examination.

Transmission, control, and treatment

Noroviruses are primarily transmitted via fecal-contaminated hands, food, water, and environmental surfaces [31-33]. In addition, a recent study confirmed that vomiting may also spread the virus through fomites and airborne droplets [34]. The primary risk factors for infection include improper food handling and contact with a person who has gastroenteritis [35]. The extremely low infectious dose of these viruses facilitates their transmission between hosts. A study published in 2008 by Teunis et al. estimated that as few as 18 virus particles would be sufficient to cause infection in 50% of exposed, susceptible subject [24]. Certain factors also promote environmental transmission; the viral RNA can persist on surfaces for at least two weeks, is resistant to some forms of chemical disinfection, and is environmentally stable at a wide range of temperatures, making it well adapted for transmission in human populations [36,37].

Although vaccines have been introduced for other enteric viruses such as rotavirus, currently no vaccine has been licensed for noroviruses. The genetic diversity of noroviruses, combined with frequent mutations within and between norovirus genotypes, have made developing a vaccine with broad protection against noroviruses challenging

[11]. An additional issue is that immunity after natural infection is often short-term, potentially as little as two months in certain individuals, suggesting that developing a vaccine with suitable longer term protection might not be cost-effective or feasible [11,38]. Despite these challenges, several candidate vaccines are presently in development. Recent efforts have utilized virus-like particle antigens and capsid proteins of noroviruses, similar to the vaccine used to prevent cervical human papilloma virus infection [39,40]. As of August 2018, eight candidate vaccines were in various stages of development, and a licensed vaccine is anticipated to be available within the next five years [39].

Since no vaccine exists at this time, interrupting transmission is the only effective strategy to preventing norovirus transmission. Good hygiene practices, as well as cleaning and disinfecting contaminated surfaces, are essential steps to prevent disease transmission. Other strategies recommended to decrease the risk of transmission include handling and preparing food safely, avoiding food preparation and caring for others while ill, and quarantining of cases [41]. However, avoiding contact with others while symptomatic, and in the days after symptoms subside, may not be a practical or sufficient strategy to control disease spread, especially considering the mean duration of viral shedding is an estimated 8 to 60 days [42].

Treatment for norovirus is non-specific and primarily consists of replenishing fluid and electrolyte losses resulting from acute vomiting and diarrhea. Because antiviral medications are not available, over-the-counter oral rehydration solutions are typically recommended as first-line therapy for cases of mild dehydration. Although cases that are more serious may require hospitalization and treatment with fluids intravenously. The

annual cost of treatment in the United States is estimated to be more than 273 million dollars among children alone, which further highlights the need for vaccine development [43].

B. Virus classification and laboratory diagnosis

Virus classification

Noroviruses are a group of non-enveloped, single-stranded RNA viruses that are 27-35 nm in diameter and belong to the *Caliciviridae* family [44]. Currently more than seven genogroups and forty genotypes have been identified (Figure 1), and three of the genogroups are known to infect humans (GI, GII, and GIV) [45,46]. The GII.4 genotype has been the most common cause of norovirus illnesses globally, but other genotypes have emerged in recent years and contribute to substantial proportions of norovirus illnesses in some areas of the world. Surveillance data of patients with acute gastroenteritis in China between 2014 and 2016 indicated that GII.17 and GII.2 genotypes have become more prevalent in certain areas of Shanghai, and GII.17 predominated at times [47]. Furthermore, new GII.4 variants have emerged every couple of years, adding to the genotype's genetic diversity and resulting in numerous epidemics [45,47]. Because noroviruses rapidly evolve over time and are genetically diverse, long-term immunity may not be possible in human populations, even after prior infection.

Diagnostic methods

Although noroviruses were first identified using immune electron microscopy, this method has a low sensitivity and is not sufficient to accurately diagnose norovirus infection [48]. Improved diagnostic methods have increased awareness of the global

burden of norovirus over the past thirty years. Because being able to detect norovirus cases is a significant public health priority in outbreak settings, rapid laboratory diagnostic methods are essential. However, the absence of an available animal cell culture model and lack of a widely available *in vitro* model have made developing diagnostic assays challenging [49].

Enzyme immunoassays/antigen detection

Several enzyme immunoassays (EIAs) are commercially available for norovirus detection, including IDEIA Norovirus and RIDASCREEN [50,51]. EIAs are a less expensive alternative to molecular detection assays for detecting norovirus antigen in clinical specimens [52]. However, viral loads and genotypes present in stool samples contribute to large variability in the sensitivities and specificities of EIAs. For example, the RIDASCREEN EIA has an estimated sensitivity of 31.6% to 92.0% and an estimated specificity of 65.3% to 100% [53-55]. Furthermore, EIAs tend to perform best when identifying an outbreak compared to individual sporadic cases of norovirus [46]. Because of the low sensitivity, the Food and Drug Administration recommends EIAs for rapid screening during outbreak settings, not to identify sporadic cases. EIAs are especially useful in outbreak settings because unlike nucleic acid amplification tests, they are relatively simple to perform in a timely manner and do not require specialized laboratory facilities [56].

Lateral-flow immunochromatographic (ICG) assays were also designed to test individual samples rapidly. Like EIAs, ICG assays have a wide range in reported sensitivities, ranging from 17.0% to 90.2%, and relatively high specificities, ranging from

87.5% to 100% [57,58]. These tests may provide an alternative to standard EIAs in point-of-care settings [56].

Molecular diagnostics

Molecular techniques are also used to detect noroviruses in clinical samples. In the 1990s, reverse transcription-polymerase chain reaction (RT-PCR) methods were developed when Jiang et al. first reported using conventional RT-PCR to detect Norwalk virus [59]. In recent years, conventional RT-PCR has become the gold standard for diagnosis of norovirus, and numerous assays have been developed [52]. Despite undergoing various improvements, conventional RT-PCR methods have a number of disadvantages, including being time-consuming and risk of sample contamination [56,60]. Consequently, most clinical virology research laboratories utilize real-time RT-PCR assays for virus detection.

Real-time RT-PCR methods significantly decrease the reaction setup time and the potential for carry-over contamination of the conventional PCR methods [56,61]. Additionally, studies have demonstrated that real-time assays have a sensitivity of approximately 5-300 genomic copies per reaction for GI and GII noroviruses [62,63]. However, primers and probes, reagents, and thermocycling conditions may also influence the sensitivity and cause substantial variability, suggesting that further research needs to be done to provide a more comprehensive analysis of using real-time assays for norovirus detection [64].

Commercial multiplex panels to diagnose infectious diseases were first created to detect respiratory viruses, but recent efforts have expanded to include gastrointestinal pathogens. The first manufactured multiplex panel for diarrheal diseases was the xTAG

Gastrointestinal Pathogen Panel, which can detect numerous pathogens including norovirus genogroups I and II [56]. Reports by the manufacturer state good sensitivities and specificities for norovirus detection, 100% and 97%, respectively [65]. Even though multiplex panels can test for numerous pathogens, they are generally time-consuming, complex, and prone to contamination. In order to combat some of these disadvantages, the BioFire FilmArray GI panel was developed. In contrast to other multiplex panels, BioFire is moderately complex, produces results in an hour, and performs all steps in a single disposable pouch [56]. In addition, an evaluation of the BioFire panel indicated a 96.2% sensitivity and 99.8% specificity for norovirus genogroups I and II [66].

C. Human challenge studies and outbreaks

Human challenge studies, also known as "controlled human infection studies" or simply "volunteer studies," involve purposefully infecting healthy volunteers with a pathogen to induce infection in order to gain insights into microbial pathogenicity, virulence factors, and host immune response [67,68]. Human challenge studies play an important role in the development of effective vaccines and antimicrobial treatments.

Results from the first norovirus human challenge study conducted by Dolin et al. were published in 1971. This study confirmed that non-bacterial gastroenteritis could be induced by orally administering stool filtrates to volunteers, supporting the hypothesis that the causative agent in the Norwalk outbreak was infectious [69]. The original Norwalk virus isolate was derived from a volunteer who developed symptoms of gastroenteritis after receiving the first passage inoculum; the resulting inoculum later became known as 8fIIa and was used in numerous subsequent challenge studies [70]. The

second Norwalk inoculum, 8fIIb, was derived from a stool sample of a volunteer who was infected with 8fIIa in a later challenge study [24,71].

D. Dose-response models

Quantitative microbial risk assessment (QMRA) is a mathematical framework used to evaluate health risks from pathogens in order to improve knowledge and management of microbial hazards [72]. One component of QMRA is dose-response assessment, which is the study of the relationship between the magnitude of exposure and the probability of infection and illness. The human body has numerous defense mechanisms in place to prevent colonization, and consequently infection, from occurring [73]. However, any single microorganism has the potential to colonize the host, given that it survives these host barriers. Using dose-response assessment, the probability of infection and illness can be modeled as a function of dose, while also including information about other host and pathogen characteristics, where available [74,75].

The assumption that any single virus particle that survives the host barriers is the lowest dose sufficient to cause infection serves as the basis for single-hit dose-response models under the independent action hypothesis [76,77]. These models treat exposure as a discrete number of organisms because microbial pathogens may cause infection at low doses, even doses consisting of merely a few virus particles. Dose is then calculated by multiplying the titer of the inoculum by the volume consumed by the study subject. Single hit models also assume that the probability of infection increases monotonically, meaning that higher doses result in a higher probability of infection. Infection may be difficult to determine in certain situations, therefore illness endpoints are sometimes used to model the probability of illness. Furthermore, these models assume that asymptomatic

infection can occur, whereas the converse (illness without infection) is not possible. The probability of illness is calculated by multiplying the conditional probability of illness given infection by the probability of infection [76].

Data from controlled human challenge studies have previously been used to estimate dose-response models for other pathogens, including *Salmonella*, *Campylobacter*, influenza, and adenovirus [78-81]. However, relatively few studies have been conducted for norovirus, though those that have mainly focus on Norwalk virus (GI.1). In 2008, Teunis et al. published the first dose-response assessment for Norwalk virus that incorporated differential host susceptibility and virus aggregation [24]. This study used a Beta Poisson model for microbial infection to describe the variation in infectivity among susceptible individuals, accounting for virus aggregation in the oral inoculum. Atmar et al. published a subsequent study of Norwalk virus in 2014 that estimated a higher median infectious dose (ID_{50}) compared to the Teunis et al. study, 1320 genome copies compared to 18-1015 genome copies, respectively [82]. A 2014 study by Messner et al. added data from four additional studies to the Teunis et al. study to calculate updated parameter estimates of the Beta Poisson dose-response model while also proposing a simpler model known as the fractional Poisson [83]. Although various mathematical models have been evaluated, no single consensus model has been recommended [84].

The Teunis et al. study used data from two clinical trials in human volunteers and did not use previous challenge study data because the estimated dose for those studies was unknown [24]. However, this study provides estimates of the GI virus concentrations (in genome copies per microliter) for two Norwalk virus inocula (8fIIa and 8fIIb) from the original qPCR data. Since these estimated concentrations are not statistically different

from other published estimates, they can be used to extend the existing model by including data from other challenge studies. Although the Messner et al. study added four additional studies to the Beta Poisson model, no studies published prior to 2008 were considered. Teunis et al. argues that the difference in aggregation state of the inocula is sufficient to explain the difference in dose-response between 8fIIa and 8fIIb, meaning that these two inocula may be considered equally infectious [24]. However, the effect of storage time on the infectivity of the inoculum is unknown. By including older studies in the model, the overall estimated infectivity of Norwalk virus may change, and older studies may exhibit a shift in dose-response compared to studies that are more recent. Understanding whether or not the concentration of virus in the inoculum or its infectivity have changed over time may be important for understanding the dose-response relationship.

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Chapter 2: Manuscript

Abstract

Noroviruses are the leading cause of acute viral gastroenteritis among all age groups worldwide. This study used both older and recent Norwalk virus (GI.1 norovirus genotype) to model the dose-response relationship for Norwalk virus infection and illness. By comparing data from all published Norwalk virus human challenge studies (N=17) to studies published in 2008 or later (N=5), we assessed whether the 8fIIa dose-response relation has shifted over time. Data from the 17 published challenge studies were used to establish a Beta Poisson dose-response model that accounts for variation in susceptibility among hosts while also accounting for the different inocula used in these studies (8fIIa, 8fIIb, or pool lot number 42399). We found no evidence to suggest a shift in the dose-response model among recent studies compared to all studies and concluded that the concentration of infectious virus in the 8fIIa inoculum has apparently persisted over time. We estimated that the average probability of infection for a single Norwalk virus particle is approximately 0.04, and our estimate of the ID_{50} of Norwalk virus is 5.65×10^3 GEC (95% CI: 687-34,231) among susceptible hosts. These results are consistent with other published estimates of Norwalk virus infectivity and pathogenicity. Future research should focus on expanding existing norovirus dose-response models to incorporate data from human challenge studies using other norovirus genotypes and outbreak data.

Background

Human noroviruses are recognized as important etiological agents of acute non-bacterial gastroenteritis infections among all age groups worldwide and are the most common cause of gastroenteritis in the United States, causing an estimated 19 to 21 million cases annually [1-3]. First recognized as a cause of acute gastroenteritis during an outbreak in Norwalk, Ohio in 1968, illness among primary cases was characterized by vomiting, diarrhea, nausea, abdominal cramps, and low-grade fever lasting 24-48 hours, on average [4]. Today noroviruses are divided into 7 genogroups (Figure 1), with genogroups I, II, and IV known to cause infection in humans [5-7]. In the U.S., outbreaks occur most frequently in long-term care facilities but have also been reported in restaurants, schools, cruise ships, and hospitals [8]. Although cases are usually self-limited, the risk of more severe illness and death is greatest among older adults, young children, and immunocompromised individuals [9,10]. Noroviruses are most commonly transmitted person-to-person, though they may also be spread by ingestion of fecal-contaminated food or water, contact with contaminated fomites, and through aerosolized particles [11,12]. These viruses are extremely infectious with high person-to-person transmission rates, posing significant challenges for infection control [13]. Adding to their infectivity, noroviruses are able to persist in water, food, and environmental surfaces for a few days up to several months after contamination occurs, and they can be shed in stool even after clinical symptoms have resolved [14-16].

The inability to propagate human noroviruses in cell culture until recently, combined with the lack of a simple sensitive assay to detect multiple strains of the virus in clinical and environmental samples, have limited norovirus research efforts and

vaccine development. Research on infectivity, virulence, and immune response has primarily used controlled studies with human volunteers, and over a dozen norovirus challenge studies have been published in the scientific literature since the 1970s. In the absence of an appropriate animal model, these controlled human challenge studies have been vital to studying norovirus pathogenicity and immunity. Previous research has indicated that certain host characteristics, such as the absence of a functional fucosyltransferase 2 (FUT2) gene, make some individuals (non-secretors) resistant to Norwalk virus (GI.1.) infection [17]. Susceptibility may also vary by norovirus genotype, further highlighting the need for multi-level models to quantify virus infectivity.

To better understand virus transmission, the relationship between the magnitude of exposure and the resulting probabilities of infection and illness must be characterized. Information about controlled norovirus exposure from human challenge studies can be used to quantify infectivity and pathogenicity across tested strains. Considering the substantial number of norovirus outbreaks worldwide, being able to quantify infectivity is of particular interest for public health in order to understand and mitigate risks associated with norovirus. Despite the available data from published challenge studies, few dose-response assessments have been performed. The first study to attempt to quantify the infectivity of norovirus was published in 2008 by Teunis et al. and focused on Norwalk virus [18]. Researchers challenged both secretor (Se+) and non-secretor (Se-) volunteers with either the aggregated 8fIIa or the dissociated 8fIIb inoculum and used a Beta Poisson model that accounted for differential host susceptibility and virus aggregation. In 2014, Messner et al. expanded on Teunis's work to produce updated parameter estimates for the Beta Poisson model and also proposed an alternative model, known as the

fractional Poisson [19]. Both of these studies assumed that differences in the dose-response curves are attributable to the aggregation state of the inoculum and concluded that the two inocula are equally infectious. These single-hit models also assume that any single microorganism that is ingested has the potential to cause infection if it survives all host barriers, assuming independent action. Other dose-response assessments have been performed since 2013 utilizing human challenge study data, as well as a study by Thebault et al. that used data from oyster-related norovirus outbreaks [20-23]. These studies support previous findings that the risk of norovirus infections is high, even at low and moderate doses.

The 8fIIa inoculum has been used in numerous challenge studies (Table 1a) and was prepared from the original Norwalk virus isolate in 1971 [24]. This inoculum has been stored in a stock suspension containing high concentrations of protein, which promoted viral aggregation [18]. The 8fIIb inoculum was prepared in 1997 from a stool sample of a volunteer infected with 8fIIa in a 1995 clinical trial [18]. Unlike the 8fIIa inoculum, 8fIIb appeared to be dissociated based on examination by electron microscopy. Studies conducted prior to 2008 have used the same 8fIIa inoculum as recent studies, yet whether the inoculum may have lost infectivity or aggregated during storage is unclear. Previous dose-response assessments have been performed using data collected in a relatively short span of time, often at a single point in time, making them unsuitable to assess if the concentration of infectious virus in the 8fIIa inoculum has declined substantially over time. Recent dose-response studies have accounted for differences in virus aggregation, but studies conducted prior to 2008 did not. No previous dose-response

assessment has utilized data collected prior to 2008, even though including data from older challenge studies may produce meaningfully different results.

In order to establish a dose-response model for Norwalk virus infection and illness, we collected data published since the initial 1972 challenge study and developed a model to study the variation in Norwalk virus dose-response by virus pathogenicity. Based on guidelines from a 2017 study comparing various proposed models for norovirus dose-response, we fit a Beta Poisson model to only the data from Norwalk virus challenge studies published since 2008 (Table 1b) [25]. We then compared this reduced model to the model that included the data from all the Norwalk virus challenge studies (Table 1a) to show that the concentration of infectious virus in the 8fIIa inoculum appears to have remained relatively stable over time. By incorporating older challenge study data in the model, this study provides updated estimates of Norwalk virus infectivity.

Methods

Challenge study data

We searched PubMed to identify relevant human challenge studies for this analysis. The search terms included “norovirus,” “Norwalk virus,” “Norwalk-like virus,” and “acute gastroenteritis” in combination with “human challenge study” or “volunteer study.” Studies were selected if they provided information on the number of study participants, the dose of inoculum each participant received, and the number of participants who were infected and/or developed subsequent symptoms of acute gastroenteritis. We excluded studies with participants who were re-challenged since some short-term immunity may exist after infection. If data were used in multiple studies, then only the most complete study was considered in the analysis.

From the selected studies, we created a database to include the following information from each study: volume of the inoculum (mL), virus genotype, total number of subjects, number of symptomatic subjects, first author, article title, journal, and year of publication, as well as the secretor status and number of infected subjects, where available. We also included information about which inoculum was used in the Norwalk virus challenge studies (8fIIa, 8fIIb, or lot number 42399) to account for differential virus concentration. We assumed that definitions of infection and illness are comparable among selected studies. Estimates for the Norwalk virus concentration in quantitative real-time PCR defined genome equivalent copies per µL (GEC/µL) for 8fIIa and 8fIIb inocula were obtained from Teunis et al. and assumed to be 3.24×10^7 and 6.92×10^5, respectively [18]. The Norwalk virus concentration of the inoculum used in the Atmar et al. study (pool lot number 42399) was assumed to be 400 GEC/µL [20]. We also included studies with subjects whose secretor status is unknown and used the number of symptomatic subjects to estimate the number of infected subjects in studies that only reported illness endpoints. Similar to previous studies, we restricted the dose-response assessment to Se+ subjects since Se- individuals have demonstrated immunity to Norwalk virus even at high doses [18,19,26].

Dose-response modeling

Compared to chemical toxins, which usually consist of many particles, relatively few pathogenic microbes—even a single infectious particle—can produce infection in human hosts [18]. Thus, we treated exposure as a discrete number of organisms: a sample of suspension with expected dose was calculated by multiplying the Norwalk virus concentration in the inocula suspension (c) by the volume ingested (V). Virus

aggregation was accounted for by using the negative binomial exposure model proposed in the 2008 Teunis et al. study [18]. We used a two-level Beta Poisson model to allow for variation in susceptibility to infection and illness within and between challenge studies. At a given dose, infected subjects were assumed to be a binomial sample of those exposed, and symptomatic subjects were treated as a binomial sample of those infected, with both binomial probabilities depending on dose. The Beta Poisson model of infection dose-response is written as

$$P_{inf}(\mathrm{cV}) = 1 - {}_1F_1(\alpha, \alpha+\beta, -\mathrm{cV}) \quad (1)$$

with the transformed parameters

$$u_1 = \frac{\alpha}{\alpha+\beta}; \qquad w_1 = \log\left(\frac{u_1}{1-u_1}\right); \qquad v_1 = \alpha+\beta; \qquad z_1 = \log(v_1) \quad (2)$$

where w_1 is a measure of infectivity (or location) and z_1 is a measure of variation in infectivity (or spread) [27]. Equation 1 above assumes that the probability of infection (p_m) given that any ingested pathogen survives all (m) host barriers to infection is Beta distributed with $p_m \sim \mathrm{Beta}(\alpha, \beta)$ and that exposure follows a Poisson distribution; α and β are the parameters that characterize infectivity.

The conditional probability of illness among infected subjects is modelled as

$$P_{\mathrm{ill|inf}}(\mathrm{cV}) = 1 - \left(1 + \frac{\mathrm{cV}}{\eta}\right)^{-r} \quad (3)$$

with transformed parameter estimates

$$u_2 = \frac{r}{r+\eta}; \qquad w_2 = \log\left(\frac{u_2}{1-u_2}\right); \qquad v_2 = r+\eta; \qquad z_2 = \log(v_2) \quad (4)$$

where as above, w_2 is a measure of location and z_2 is a measure of spread [28]. The parameters r and η are the parameters that characterize illness.

Parameters were estimated by host susceptibility for secretor-positive subjects. The parameters for location of infectivity (w_1) and pathogenicity (w_2) were given normal priors with the effects of secretor status as

$$w_1(\text{Se}) \sim N(\mu_{w,1}, \tau_{w,1}); \qquad w_2(\text{Se}) \sim N(\mu_{w,2}, \tau_{w,2}) \tag{5}$$

of fixed precision $\tau_{w,1}$ and $\tau_{w,2}$, respectively ($\tau \equiv 1/\sigma^2$). The parameters for spread of infectivity (z_1) and pathogenicity (z_2) were given normal priors

$$z_1(\text{Se}) \sim N(\mu_{z,1}, \tau_{z,1}); \qquad z_2(\text{Se}) \sim N(\mu_{z,2}, \tau_{z,2}) \tag{6}$$

with broad priors ($\tau_{w,1} = \tau_{w,2} = 0.05$).

Variation in infectivity for host susceptibility was given a hyperprior $\mu_1 \sim N(0, 0.01)$, and variation in pathogenicity for host susceptibility was assumed $\mu_2 \sim N(0, 0.01)$. The remaining precision of w_1 and w_2 within host was given as $\tau_w = 1$. Priors for z_1 and z_2 were $N(0, 1)$.

All analyses were performed using a model adapted from the 2018 Teunis et al. study of *Campylobacter jejuni* in JAGS (version 4.3.0) and R (version 3.5.1) [29]. Source code is provided in the Appendix. Four parallel chains were run. After a burn-in of 10^3 iterations, the model was run for 10^5 iterations, and a posterior sample size of $4 \times 5000 = 20{,}000$ was used. Convergence was checked by inspection of posteriors. Differences between dose-response models were evaluated by comparing (posterior predictive) Monte Carlo samples of doses required for 50% infection and illness using an approach proposed by Gelman et al. [30].

Results

Table 1a provides information about the 17 challenge studies included in this analysis. Studies in Table 1a were included in the full analysis, while 5 studies in Table 1b were included in the analysis of only recent studies. Both of these analyses included data for all three Norwalk virus inocula. Figure 2 shows dose-response relations for infection in all Norwalk virus human challenge studies (Figure 2a) and in challenge studies published in 2008 or later (Figure 2b). The similar shapes of the curves suggest that prolonged storage of the 8fIIa inoculum has not meaningfully changed the concentration of infectious virus over time. The differences between the dose-response models based on the Monte Carlo samples of doses required for 50% infection further support this finding, with 50.7% of the differences greater than 0. This indicates that the samples from newer studies have a similar distribution as the samples from all included studies, meaning that there is no apparent shift in the dose-response curves. Figure 2(a) shows a predicted generalized dose-response relation for all studies included in the analysis, which may be interpreted as the probability of infection as a function of dose, for Norwalk virus among Se+ subjects.

In Figure 3, dose-response relations are shown for the unconditional probability of illness, calculated as $P_{\text{inf}}(\text{cV}) \times P_{\text{ill}|\text{inf}}(\text{cV})$, for all Norwalk virus human challenge studies selected (a) and for challenge studies published in 2008 or later (b). Again, the similar shapes of the dose-response curves suggest that the virulence of the 8fIIa inoculum has persisted over time. Using the Monte Carlo samples to compare the illness dose-response models for all studies compared to newer studies, we found that 49.5% of the differences were greater than 0, suggesting that the two models are not meaningfully different.

Table 2 lists quantiles of the doses required for 25% infection and 25% illness, as well as doses for 1% infection and 1% illness. Similarly, Figure 4 shows boxplots of the estimated doses for 1% and 50% infection or illness. These estimates are commonly referred to as ID_{01} and ID_{50} and represent the expected doses necessary to cause infection or illness in 1% and 50% of susceptible individuals; to distinguish between infection and illness, we refer to these estimates as $InfD_{01}$ and $InfD_{50}$. The dose response relation among secretor-positive subjects has an estimated $InfD_{01}$ of 0.2 GEC (Table 2) and an estimated $InfD_{50}$ of 5.65×10^3 GEC. An additional illustration of the infectivity and pathogenicity is given in Table 3, which shows the probability of infection and illness at a mean dose of 1 genome equivalent copy (GEC) of Norwalk virus. We found that the median probabilities of infection and illness among susceptible hosts exposed to a mean dose of 1 GEC of Norwalk virus were approximately 0.043 and 0.0029, respectively.

Tables 4 provides estimates of the dose-response parameters for infection and illness among secretors for the models in Equation 1 and Equation 3. The parameters α and β characterize infectivity, while r and η are the parameters that characterize illness. Table 5 provides transformed dose-response parameters, with w_1 and w_2 representing measures of the central tendency (or location) of infectivity, and z_1 and z_2 representing measures of the variation in infectivity (or spread).

Discussion

Noroviruses remain a significant cause of acute gastroenteritis globally each year. Dose-response information is useful for characterizing the health hazards associated with noroviruses and for developing quantitative risk assessments. This is the first known

study to assess the change in concentration of infectious virus in the 8fIIa Norwalk virus inoculum over time. By including data from Norwalk virus challenge studies published both prior to 2008 and more recently, we established a Beta Poisson model for Norwalk virus infectivity and pathogenicity using a model that has previously been used for *Campylobacter jejuni* [29]. Consistent with other Norwalk virus studies, we showed that Norwalk virus is highly infectious and estimate that the median probability of infection among susceptible individuals at a mean dose of 1 GEC is approximately 0.04. Adding data from older challenge studies that exclusively used the 8fIIa inoculum did not meaningfully change the estimated infectivity or pathogenicity of Norwalk virus.

Our study estimated that the $InfD_{50}$ among susceptible hosts was 5.65×10^3 GEC (95% CI: 690-34,000). Atmar et al. reported an estimated ID_{50} of 1,320-2,800 GEC, depending on histo-blood group [18,20]. Among all secretor-positive individuals in the Atmar et al. study, the 95% CI for the ID_{50} was 290-25,000 GEC, which is consistent with our result. Using the Fractional Poisson model and including data from challenge studies using all three inocula, Messner et al. estimated that the mean number of viruses per aggregate is approximately 1,106 (95% CI: 399-2,428) [19]. It is important to note that ID_{50} studies are prone to substantial statistical uncertainty in outcomes because of a variety of factors, including the relatively small number of subjects examined in challenge studies, differential host susceptibility, and viral aggregation of inocula. This is evident when examining the 95% confidence intervals for these estimates, which can be quite large. Our estimate of the $InfD_{50}$ among susceptible hosts appeared somewhat higher than other published estimates, but examination of the confidence intervals suggest that the results are similar.

This study is not without limitations. Although utilizing data from older challenge studies is valuable, these studies typically used illness as an endpoint rather than infection. Consequently, we may underestimate Norwalk virus infectivity because individuals with asymptomatic infections would be excluded from the analysis. For older studies that reported both infection and illness, infection was determined using enzyme immunoassay methods because PCR methods were not available until the 1990s [31-34]. In contrast to molecular techniques, antigen detection methods tend to be less sensitive and are no longer recommended for use in identifying individual sporadic cases [35-37]. Older studies also did not report secretor status for subjects, thus these studies may have included some secretor-negative individuals who would not be susceptible to Norwalk virus infection.

Another limitation is that the correction for virus aggregation is only applied to the 8fIIa data from the 2008 Teunis et al. study, and not to any of the older studies using that inoculum. Based on this limitation, our analysis does not contradict the conclusion that the 8fIIa inoculum has not deteriorated due to virus inactivation over the time it has been studied. Human challenge studies serve as a crude instrument to characterize infectivity and pathogenicity of viruses compared to methods used for bacteria, but these studies are the best tool available until cell culture models become widely available for human noroviruses. Additionally, we used a previously published estimate for the number of genome copies/µL contained in each inoculum, although other estimates have been published. The estimates produced in the 2008 Teunis et al. study indicate that apparent differences in concentration for the same inoculum are likely too small to substantially influence the outcomes, but this assumption was not assessed in this study.

We tested the hypothesis that the concentration of infectious virus in the 8fIIa inoculum has not meaningfully deteriorated over time by examining whether a shift in dose-response is consistent with the observed outcomes. Alternatively, we could have assumed that the concentration of infectious virus has declined over time and estimated the virus concentration at various time points using the challenge data, similar to the Teunis et al. study of *Campylobacter jejuni* [29].

While human challenge studies often involve exposing subjects to relatively high doses, exposure to pathogens usually occurs at low doses in natural settings. Therefore, being able to estimate infection risk at low doses is critical. Predictions of infectivity and illness at low doses play a significant role in quantitative microbial risk assessment, particularly in areas related to food and water quality. Additional human challenge study data and outbreak data can be used to increase the precision and validity of these estimates, especially at low doses. Only one previously published study has analyzed data from norovirus outbreaks associated with oyster consumption, and no published study has jointly analyzed both human challenge and outbreak data [23]. When the virus concentration and dose can be reliably estimated using quantitative real-time PCR in outbreak settings, these data can be used to assess how well current models fit low-dose estimates.

Although this study focused on Norwalk virus, other norovirus genotypes circulate in humans and are therefore relevant in dose-response models for risk assessment. While individuals without a functional fucosyltransferase (FUT2) enzyme have demonstrated lack of susceptibility to Norwalk and GII.4 virus infection, secretor status does not seem to provide protection against some other commonly seen norovirus

genotypes such as Snow Mountain virus (GII.2) [17,26,38]. Moreover, GII.4 viruses have caused an estimated 46.8-70.4% of outbreaks in the United States in recent years, while GII.17 and GII.2 genotypes have predominated during certain time periods in China [39,40]. Given the emergence of new genotypes and the genetic diversity of noroviruses as a whole, expanding existing norovirus dose-response models to incorporate other genotypes is a crucial next step in increasing understanding of norovirus infectivity and pathogenicity.

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Figures and Tables

Figure 1. Classification of noroviruses into 7 genogroups (GI to GVII) based on amino acid sequence diversity in the complete VP1 capsid protein [7].

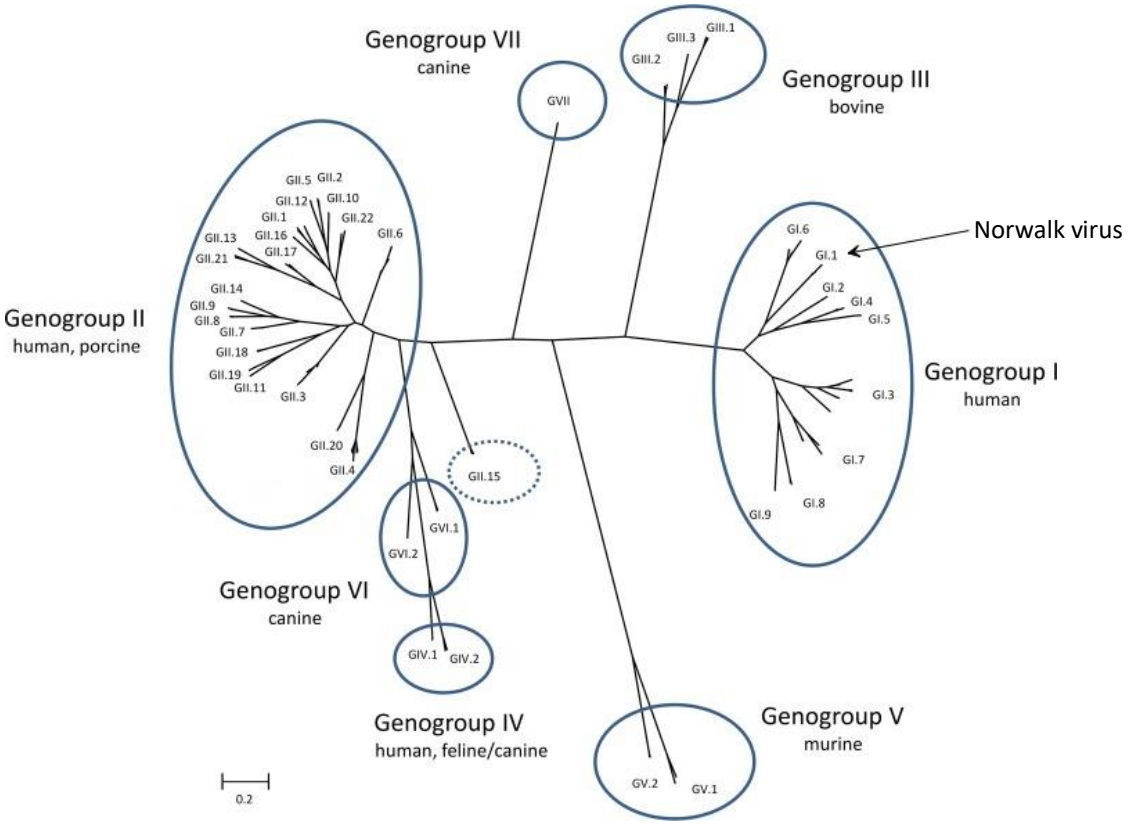
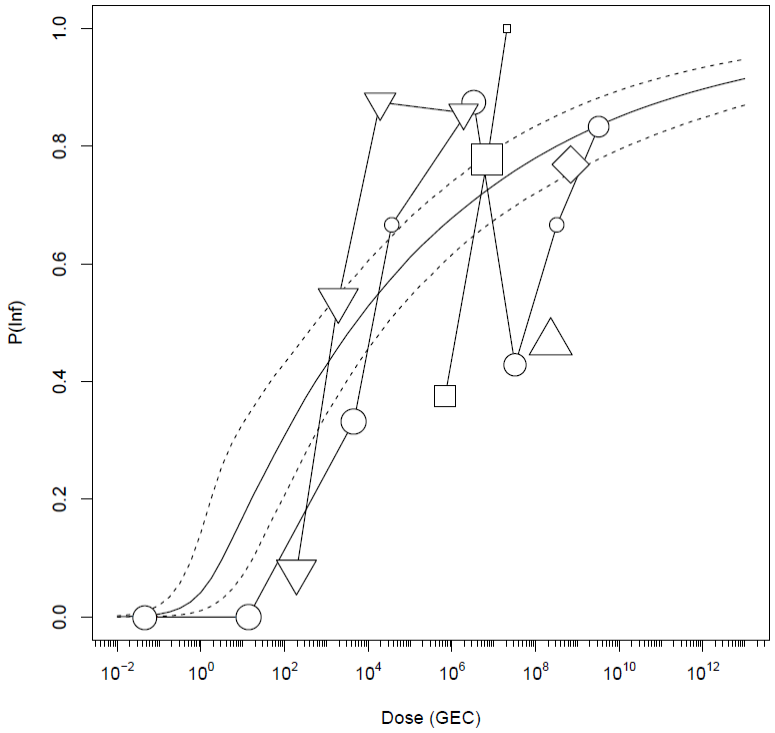
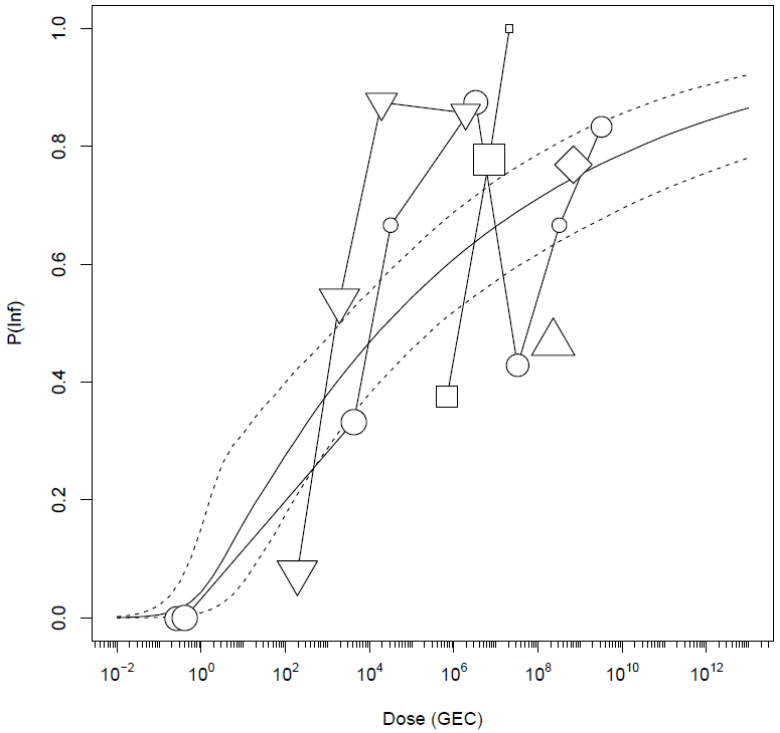


Figure 2. Dose-response model for Norwalk virus infection among secretors using data from all published human challenge studies included in the analysis (N=17) (a) and for studies published in 2008 or later (N=5) (b). Shown are median and 95% predicted intervals for probability of infection as a function of dose. Observed fractions are shown as a bubble chart, and sizes of the symbols represent the fractions of either the observed number of infected or ill proportional to the number exposed at the challenge doses. Different symbols indicate different studies; data from the same study are connected.

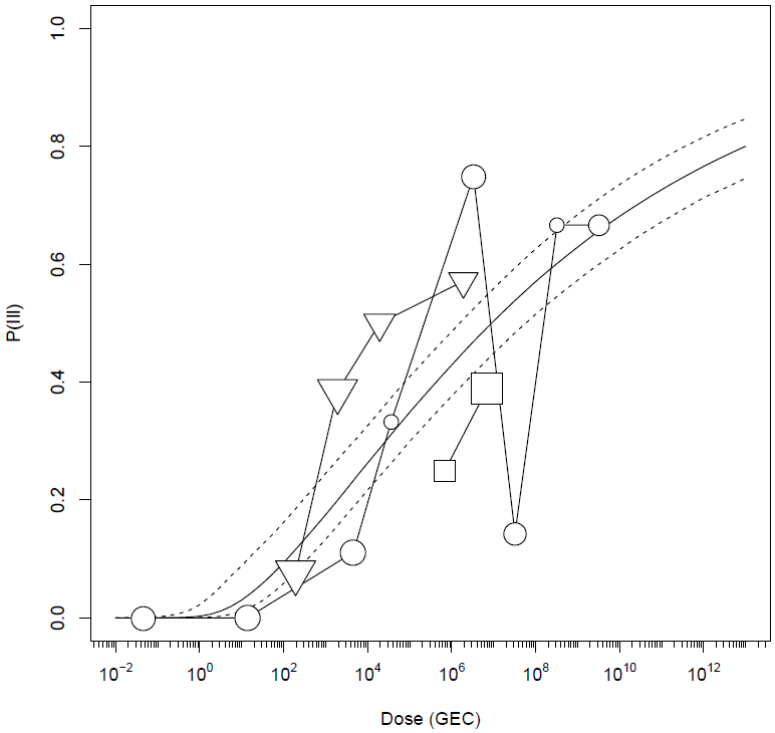


(a) All studies (N=636 subjects)

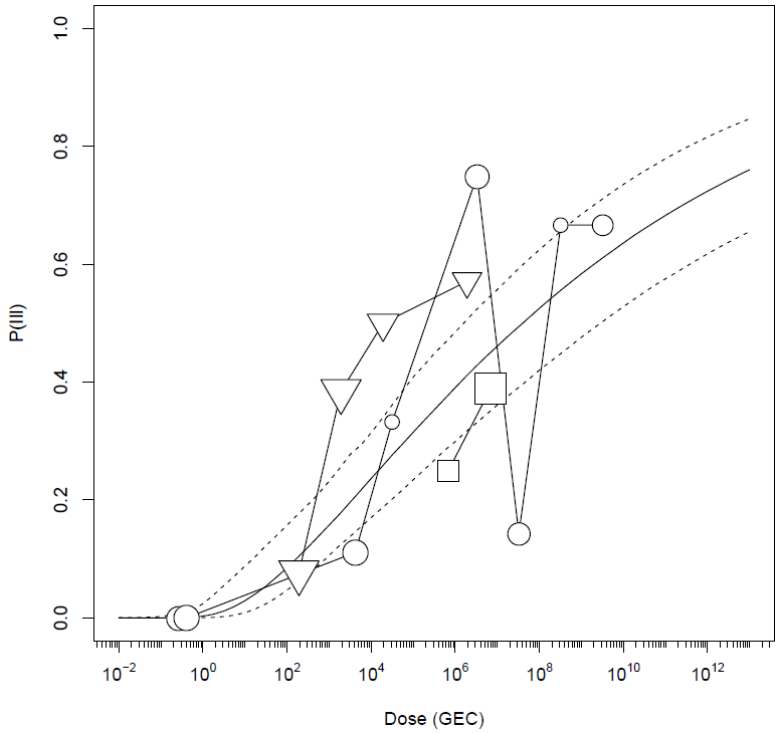


(b) Studies published in 2008 or later (N=342 subjects)

Figure 3. Dose-response model for Norwalk virus illness among secretors using data from all published human challenge studies included in the analysis (N=17) (a) and for studies published in 2008 or later (N=5) (b). Shown are median and 95% predicted intervals for probability of illness as a function of dose. Observed fractions are shown as a bubble chart, and sizes of the symbols represent the fractions of either the observed number of infected or ill proportional to the number exposed at the challenge doses. Different symbols indicate different studies; data from the same study are connected.

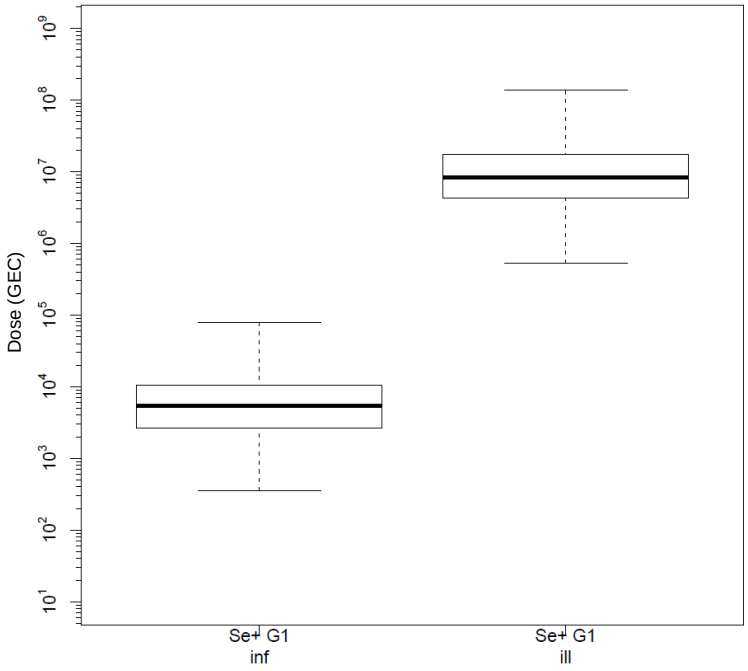


(a) All studies (N=636 subjects)

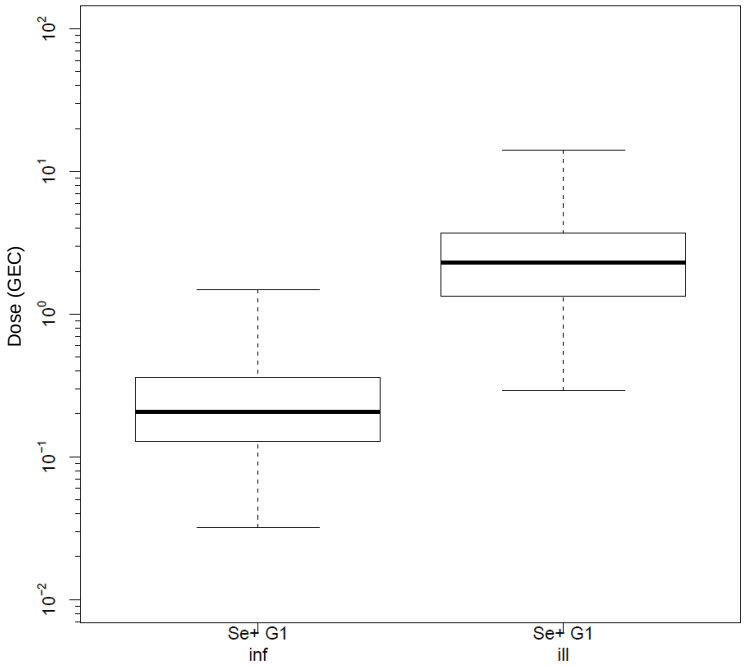


(b) Studies published in 2008 or later (N=342 subjects)

Figure 4. Dose in genome equivalent copies (GEC) required for 50% (a) and 1% (b) probability of infection and illness, respectively, among secretors from all published human challenge studies of Norwalk virus with known secretor status (N=5).



(a) 50% Dose



(b) 1% Dose

Table 1a. Norwalk virus challenge study data used in the full dose-response model.

		Volume	Titer	Dose	Non-secretors (Se-)			Secretors (Se+)			Unknown Se		
Reference	Inoculum	(µL)	(GEC/µL)	(GEC)	Exposed	Infected	Ill	Exposed	Infected	Ill	Exposed	Infected	Ill
Atmar et al. (2014)	Lot # 42399	4.80×10^{-1}	4.00×10^{2}	1.92×10^{2}	3	0	0	13	1	1			
		4.80×10^{0}	4.00×10^{2}	1.92×10^{3}	1	0	0	13	7	5			
		4.80×10^{1}	4.00×10^{2}	1.92×10^{4}	2	0	0	8	7	4			
		4.80×10^{3}	4.00×10^{2}	1.92×10^{6}	2	0	0	7	6	4			
Leon et al. (2011)	8fIIb	3.33×10^{2}	6.92×10^{5}	6.91×10^{5}				15	7	NA			
Seitz et al. (2011)	8fIIb	1.00×10^{2}	6.92×10^{5}	6.92×10^{7}				13	10	NA			
Teunis et al. (2008)	8fIIa	1.00×10^{-6}	3.24×10^{7}	3.24×10^{1}	2	0	0	8	0	0			
		1.00×10^{-5}	3.24×10^{7}	3.24×10^{2}	2	0	0	9	0	0			
		1.00×10^{-4}	3.24×10^{7}	3.24×10^{3}	6	0	0	9	3	1			
		1.00×10^{-3}	3.24×10^{7}	3.24×10^{4}	1	0	0	3	2	1			
		1.00×10^{-1}	3.24×10^{7}	3.24×10^{6}	2	0	0	8	7	6			
		1.00×10^{0}	3.24×10^{7}	3.24×10^{7}	3	0	0	7	3	1			
		1.00×10^{1}	3.24×10^{7}	3.24×10^{8}	2	0	0	3	2	2			
		1.00×10^{2}	3.24×10^{7}	3.24×10^{9}	4	0	0	6	5	4			
Teunis et al. (2008)	8fIIb	1.00×10^{0}	6.92×10^{5}	6.92×10^{5}	2	0	0	8	3	2			
		1.00×10^{1}	6.92×10^{5}	6.92×10^{6}	4	0	0	18	14	7			
		3.00×10^{1}	6.92×10^{5}	2.08×10^{7}	0	0	0	1	1	NA			
Graham et al. (1994)	8fIIa	2.00×10^{1}	3.24×10^{7}	6.48×10^{8}							50	41	28
Johnson et al. (1990)	8fIIa	2.00×10^{1}	3.24×10^{7}	6.48×10^{8}							42	29	23
Keswick et al. (1985)	8fIIa	9.00×10^{1}	3.24×10^{7}	2.92×10^{9}							16	14	11
Meeroff et al. (1985)	8fIIa	5.00×10^{2}	3.24×10^{7}	1.62×10^{10}							7	NA	4
Steinhoff et al. (1980)	8fIIa	5.00×10^{1}	3.24×10^{7}	1.62×10^{9}							59	40	34
Parrino et al. (1977)	8fIIa	3.00×10^{3}	3.24×10^{7}	9.72×10^{10}							12	NA	6

Levy et al. (1976)	8fIIa	4.00×10^{3}	3.24×10^{7}	1.31×10^{11}	16	NA	11
Widerlite et al. (1975)	8fIIa	3.00×10^{3}	3.24×10^{7}	9.72×10^{10}	15	NA	9
Wyatt et al. (1974)	8fIIa	4.00×10^{3}	3.24×10^{7}	1.31×10^{11}	36	NA	19
Agnus et al. (1973)	8fIIa	5.00×10^{3}	3.24×10^{7}	1.62×10^{11}	7	NA	4
Schreiber et al. (1973)	8fIIa	3.00×10^{3}	3.24×10^{7}	9.72×10^{10}	15	NA	12
Dolin et al. (1972)	8fIIa	5.00×10^{3}	3.24×10^{7}	1.62×10^{11}	19	NA	7

Table 1b. Norwalk virus challenge study data used in the reduced dose-response model, restricted to studies published in 2008 or later.

		Volume	Titer	Dose	Non-secretors (Se-)			Secretors (Se+)		
Reference	Inoculum	(µL)	(GEC/µL)	(GEC)	Exposed	Infected	Ill	Exposed	Infected	Ill
Atmar et al. (2014)	Lot # 42399	4.80×10^{-1}	4.00×10^{2}	1.92×10^{2}	3	0	0	13	1	1
		4.80×10^{0}	4.00×10^{2}	1.92×10^{3}	1	0	0	13	7	5
		4.80×10^{1}	4.00×10^{2}	1.92×10^{4}	2	0	0	8	7	4
		4.80×10^{3}	4.00×10^{2}	1.92×10^{6}	2	0	0	7	6	4
Leon et al. (2011)	8fIIb	9.99×10^{-1}	6.92×10^{5}	6.91×10^{5}				15	7	NA
Seitz et al. (2011)	8fIIb	1.00×10^{2}	6.92×10^{5}	6.92×10^{7}				13	10	NA
Teunis et al. (2008)	8fIIa	1.00×10^{-6}	3.24×10^{7}	3.24×10^{1}	2	0	0	8	0	0
		1.00×10^{-5}	3.24×10^{7}	3.24×10^{2}	2	0	0	9	0	0
		1.00×10^{-4}	3.24×10^{7}	3.24×10^{3}	6	0	0	9	3	1
		1.00×10^{-3}	3.24×10^{7}	3.24×10^{4}	1	0	0	3	2	1
		1.00×10^{-1}	3.24×10^{7}	3.24×10^{6}	2	0	0	8	7	6
		1.00×10^{0}	3.24×10^{7}	3.24×10^{7}	3	0	0	7	3	1
		1.00×10^{1}	3.24×10^{7}	3.24×10^{8}	2	0	0	3	2	2
		1.00×10^{2}	3.24×10^{7}	3.24×10^{9}	4	0	0	6	5	4
Teunis et al. (2008)	8fIIb	1.00×10^{0}	6.92×10^{5}	6.92×10^{5}	2	0	0	8	3	2
		1.00×10^{1}	6.92×10^{5}	6.92×10^{6}	4	0	0	18	14	7
		3.00×10^{1}	6.92×10^{5}	2.08×10^{7}	0	0	0	1	1	NA

Table 2. Doses in genome equivalent copies (GEC) required for 25% infection (InfD25), 25% illness (IllD25), 1% infection (InfD01), and 1% illness (IllD01) among secretors.

$InfD_{25}$			$IllD_{25}$		
Median	2.5%	97.5%	Median	2.5%	97.5%
4.44×10^{1}	4.13×10^{0}	2.27×10^{2}	7.28×10^{3}	5.84×10^{2}	2.82×10^{4}

$InfD_{01}$			$IllD_{01}$		
Median	2.5%	97.5%	Median	2.5%	97.5%
2.17×10^{-1}	5.85×10^{-2}	1.18×10^{0}	2.77×10^{0}	3.10×10^{-1}	8.97×10^{0}

Table 3. Probability of infection and illness of Norwalk virus at mean dose of 1 genome equivalent copy (GEC) among secretors.

Pinf(1)			Pill(1)		
Median	2.5%	97.5%	Median	2.5%	97.5%
3.84×10^{-2}	8.58×10^{-3}	1.25×10^{-1}	2.85×10^{-3}	5.05×10^{-4}	3.04×10^{-2}

Table 4. Dose-response parameters for infection (α, β) and illness (r, η) among secretors.

Parameter*	Median	2.5%	97.5%
α	8.40×10^{-2}	6.49×10^{-2}	1.02×10^{-1}
β	1.68×10^{0}	3.32×10^{-1}	9.52×10^{0}
r	6.74×10^{-2}	4.18×10^{-2}	8.31×10^{-2}
η	4.14×10^{-1}	2.10×10^{-6}	2.81×10^{0}

*α and β are the parameters that characterize infectivity; r and η are the parameters that characterize illness

Table 5. Transformed dose-response parameters for infection (w_1‡, z_1*) and illness (w_2‡, z_2*) among secretors.

Infection	mean (w_1)	mean (z_1)	var(w_1)	cov(w_1, z_1)	var(z_1)
	-3.046	0.619	0.650	-0.668	0.695
Illness	mean (w_2)	mean (z_2)	var(w_2)	cov(w_2, z_2)	var(z_2)
	-0.694	-0.923	14.633	-3.434	1.427

‡ w_1 and w_2 are measures of the central tendency (or location of) infectivity
* z_1 and z_2 are measures of variation in infectivity (or spread)

Chapter 3: Public Health Implications and Future Research

Noroviruses are recognized as the leading cause of acute gastroenteritis in the United States and the most common cause of epidemic diarrheal disease worldwide [1]. According to the Centers for Disease Control and Prevention, noroviruses cause an estimated 19 to 21 million illnesses in the U.S. and an estimated 685 million cases globally each year [2]. Although disease is usually mild and self-limited, an estimated 212,000 norovirus deaths occur annually worldwide, with approximately 570 to 800 of those deaths occurring in the United States [1,2].

Reductions in global diarrheal deaths since the 1990s, particularly among children under the age of 5, are largely attributable to improvements in water, sanitation, and hygiene. The introduction of a vaccine for rotavirus in 2006 has likewise resulted in a 58% to 90% reduction in rotavirus cases [3]. Despite these successes, prevention and control of norovirus infections remains a global challenge. Norovirus infection causes approximately 18% of diarrheal diseases worldwide, with high-, middle-, and low-income settings all experiencing similar disease incidences [4]. In the absence of an available norovirus vaccine, efforts to reduce transmission have primarily focused on understanding and preventing disease transmission. Noroviruses are highly infectious, with exposure to a small number of virus particles associated with high rates of infection in volunteer studies [5-7]. A large number of foodborne outbreaks worldwide are attributable to norovirus, posing a significant public health challenge and highlighting the need to quantify norovirus infectivity and pathogenicity.

Quantitative microbial risk assessment (QMRA) is a method for estimating health risks associated with exposure to infectious microorganisms, especially those in food and

water [8]. Dose-response assessment is a critical step in any QMRA and involves quantifying the relationship between the magnitude of exposure to a pathogen and the probability of infection and illness [9]. Information from dose-response models and QMRAs can be used to increase understanding of pathogen transmission, develop plans to mitigate and prevent exposure, and implement policies to decrease the overall disease burden.

As the first identified norovirus genotype, Norwalk virus (GI.1) has previously been studied in human challenge studies to understand norovirus infectivity. This study used data from published human challenge studies of Norwalk virus to establish a Beta Poisson dose-response model for infection and illness. Our model was adapted from a 2018 *Campylobacter jejuni* study and accounted for virus aggregation, similar to previous Norwalk virus dose-response assessments [5,10,11]. Although alternative models have been used for dose-response assessment, no single consensus model is currently recommended [12,13]. Consequently, it may be worthwhile to fit multiple models to assess similarities and differences between models.

Unlike previous dose-response studies of Norwalk virus, our study was also able to assess changes in concentration of infectious virus in the 8fIIa inoculum used in volunteer studies by including data from both early and recent challenge studies. This study focused on Norwalk virus challenge studies, but challenge studies of other norovirus genotypes, including GII.4, Hawaii, and Snow Mountain viruses, have also been published [7,14-17]. The dose-response relation for infection has previously been shown to depend on pathogen strain for other pathogens, thus incorporating studies of additional norovirus genotypes into future models will be important [11,18]. Host

susceptibility also varies by genotype, therefore estimates of infectivity and pathogenicity among secretor-negative (Se-) individuals can only be calculated by including genotypes for which Se- individuals are susceptible, such as Snow Mountain virus. Future dose-response assessments should include additional norovirus genotypes, especially GII.4 viruses considering they have been the predominant genotype in the United States in recent years [19].

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Appendix

R Code

```
## Human volunteer data

# Teunis et al. (2008) Norwalk virus: How infectious is it?
# Journal of Medical Virology 80(8):1468-1476
challenge.1 <- list(strain="G1.1-8FIIa",
                    se=c(rep(0,2),rep(1,8),rep(0,2),rep(1,9),
                         rep(0,6),rep(1,9),rep(0,1),rep(1,3),
                         rep(0,2),rep(1,8),rep(0,3),rep(1,7),
                         rep(0,2),rep(1,3),rep(0,4),rep(1,6)),
                    qty=c(rep(1e-6,10),rep(1e-5,11),
                          rep(1e-4,15),rep(1e-3,4),
                          rep(1e-1,10),rep(1e0,10),
                          rep(1e1,5),rep(1e2,10)), # aggregated
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,75),
                    infec=c(rep(0,2),rep(0,8),rep(0,2),rep(0,9),
                            rep(0,6),rep(0,6),rep(1,3),rep(0,1),
                            rep(0,1),rep(1,2),rep(0,2),rep(0,1),
                            rep(1,7),rep(0,3),rep(0,4),rep(1,3),
                            rep(0,2),rep(0,1),rep(1,2),rep(0,4),
                            rep(0,1),rep(1,5)),
                    sympt=c(rep(0,2),rep(0,8),rep(0,2),rep(0,9),
                            rep(0,6),rep(0,8),rep(1,1),rep(0,1),
                            rep(0,2),rep(1,1),rep(0,2),rep(0,2),
                            rep(1,6),rep(0,3),rep(0,6),rep(1,1),
                            rep(0,2),rep(0,1),rep(1,2),rep(0,4),
```

```
                            rep(0,2),rep(1,4)));
challenge.2 <- list(strain="G1.1-8FIIb",
                    se=c(rep(0,2),rep(1,8),rep(0,4),rep(1,18),
                         rep(1,1)),
                    qty=c(rep(1e0,10),rep(1e1,22),
                          rep(3e1,1)), # disaggregated
                    qty.rn=c(NA,NA),
                    conc.g1=c(6.92e5,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,33),
                    infec=c(rep(0,2),rep(0,5),rep(1,3),rep(0,4),
                            rep(0,4),rep(1,14),rep(1,1)),
                    sympt=c(rep(0,2),rep(0,6),rep(1,2),rep(0,4),
                            rep(0,11),rep(1,7),rep(NA,1)));
# Seitz et al. 2011: Norovirus infectivity in humans and
persistence
# in water.
# Applied and Environmental Microbiology 77(19):6884-6888.
challenge.3 <- list(strain="G1.1-8FIIb",
                    se=c(rep(1,13)),
                    qty=c(rep(1000,13)),
                    qty.rn=c(NA,NA),
                    conc.g1=c(6.92e5,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=c(rep(1,13)),
                    infec=c(rep(0,3),rep(1,10)),
                    sympt=c(rep(0,3),rep(NA,10)));
# Leon et al. 2011: Randomized, Double-Blinded Clinical Trial for
Human
# Norovirus Inactivation in Oysters by High Hydrostatic Pressure
Processing.
# Applied and Environmental Microbiology 77(15):5476-5482.
challenge.4 <- list(strain="G1.1-8FIIb",
```

```
                    se=c(rep(1,15)),
                    qty=c(rep(333,15)),
                    qty.rn=c(NA,NA),
                    conc.g1=c(6.92e5,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=c(rep(1,15)),
                    infec=c(rep(0,8),rep(1,7)),
                    sympt=c(rep(0,8),rep(NA,7)));
# Atmar et al (2014) Determination of the 50% human infectious
dose
# for Norwalk virus.
# Journal of Infectious Diseases 209(7):1016-1022
challenge.5 <- list(strain="G1.1", # this is not 8FIIb
                    se=c(rep(0,2),rep(1,7),rep(0,2),rep(1,8),
                         rep(0,1),rep(1,13),rep(0,3),rep(1,13)),
                    qty=c(rep(4800,9),rep(48,10),rep(4.8,14),
                          rep(0.48,16)),
                    qty.rn=c(NA,NA),
                    conc.g1=c(400,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,49),
                    infec=c(rep(0,2),rep(0,1),rep(1,6),
                            rep(0,2),rep(0,1),rep(1,7),
                            rep(0,1),rep(0,6),rep(1,7),
                            rep(0,3),rep(0,12),rep(1,1)),
                    sympt=c(rep(0,2),rep(0,3),rep(1,4),
                            rep(0,2),rep(0,4),rep(1,4),
                            rep(0,1),rep(0,8),rep(1,5),
                            rep(0,3),rep(0,12),rep(1,1)));
# Graham et al (1994) Norwalk Virus Infection of Volunteers: New
# Insights Based on Improved Assays.
# Journal of Infectious Diseases 170(1):34-43
```

```
challenge.6 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,50),
                    qty=rep(20,50),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,50),
                    infec=c(rep(0,9),rep(1,41)),
                    sympt=c(rep(0,9),rep(0,13),rep(1,28)));
# Johnson et al (1990) Multiple-Challenge Study of Host
Susceptibility
# to Norwalk Gastroenteritis in US Adults.
# Journal of Infectious Diseases 161(1):18-21
challenge.7 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,42),
                    qty=rep(20,42),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,42),
                    infec=c(rep(1,29),rep(0,13)),
                    sympt=c(rep(1,23),rep(0,19)));
# Steinhoff et al (1980) Bismuth subsalicylate therapy of
# viral gastroenteritis.
# Gastroenterology 78(6):1495-1499
challenge.8 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,59),
                    qty=rep(50,59),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,59),
```

```
                    infec=c(rep(1,40),rep(0,19)),
                    sympt=c(rep(1,34),rep(0,25)));
# Keswick et al (1985) Inactivation of Norwalk Virus in
# Drinking Water by Chlorine.
# Applied and Environmental Microbiology 50(2):261-264
challenge.9 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,16),
                    qty=rep(90,16),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,16),
                    infec=c(rep(1,14),rep(0,2)),
                    sympt=c(rep(1,11),rep(0,5)));
# Meeroff et al (1985) Abnormal Gastric Motor Function in
# Viral Gastroenteritis.
# Annals of Internal Medicine 92(3):370-373
challenge.10 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,7),
                    qty=rep(500,7),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),# if not 8FIIa
then what?
                    conc.g2=rep(NA,4),
                    expos=rep(1,7),
                    infec=rep(NA,7),
                    sympt=c(rep(1,4),rep(0,3)));
# Parrino et al (1977) Clinical Immunity in Acute Gastroenteritis
# Caused by Norwalk Agent.
# New England Journal of Medicine 297(2):86-89
challenge.11 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,12),
```

```
                    qty=rep(3000,12),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,12),
                    infec=rep(NA,12),
                    sympt=c(rep(1,6),rep(0,6)));
# Widerlite et al (1975) Structure of the gastric mucosa in
# acute infectious bacterial gastroenteritis.
# Gastroenterology 68(3):425-430
challenge.12 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,15),
                    qty=rep(3000,15),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,15),
                    infec=rep(NA,15),
                    sympt=c(rep(1,9),rep(0,6)));
# Schreiber et al (1973) The Mucosal Lesion of the Proximal Small
# Intestine in Acute Infectious Nonbacterial Gastroenteritis.
# New England Journal of Medicine 288(25):1318-1323
challenge.13 <- list(strain="G1.1-8FIIa",
                     se=rep(NA,15),
                     qty=rep(3000,15),
                     qty.rn=c(NA,NA),
                     conc.g1=c(3.24e7,rep(NA,3)), # see Meeroff
et al (1985)
                     conc.g2=rep(NA,4),
                     expos=rep(1,15),
                     infec=rep(NA,15),
                     sympt=c(rep(1,12),rep(0,3)));
```

```
# Levy et al (1976) Jejunal Adenylate Cyclase Activity in
# Human Subjects During Viral Gastroenteritis.
# Gastroenterology 70(3):321-325
challenge.14 <- list(strain="G1.1-8FIIa",
                     se=rep(NA,16),
                     qty=rep(4000,16),
                     qty.rn=c(NA,NA),
                     conc.g1=c(3.24e7,rep(NA,3)),
                     conc.g2=rep(NA,4),
                     expos=rep(1,16),
                     infec=rep(NA,16),
                     sympt=c(rep(1,11),rep(0,5)));
# Wyatt et al (1974) Comparison of Three Agents of Acute
Infectious
# Nonbacterial Gastroenteritis by Cross-Challenge in Volunteers.
# Journal of Infectious Diseases 70(3):321-325
challenge.15 <- list(strain="G1.1-8FIIa",
                     se=rep(NA,36),
                     qty=rep(4000,36),
                     qty.rn=c(NA,NA),
                     conc.g1=c(3.24e7,rep(NA,3)),
                     conc.g2=rep(NA,4),
                     expos=rep(1,36),
                     infec=rep(NA,36),
                     sympt=c(rep(1,19),rep(0,17)));
# Agnus et al (1973) Acute Infectious Nonbacterial
# Gastroenteritis: Intestinal Histopathology.
# Annals of Internal Medicine 79(1):18-25
challenge.16 <- list(strain="G1.1-8FIIa",
                     se=rep(NA,7),
                     qty=rep(5000,7),
                     qty.rn=c(NA,NA),
```

```
                    conc.g1=c(3.24e7,rep(NA,3)), # see Meeroff
et al (1985)
                    conc.g2=rep(NA,4),
                    expos=rep(1,7),
                    infec=rep(NA,7),
                    sympt=c(rep(1,4),rep(0,3)));
# Dolin et al (1972) Biological properties of Norwalk agent of
# acute infectious nonbacterial gastroenteritis.
# Experimental Biology and Medicine 140(2):578-583
challenge.17 <- list(strain="G1.1-8FIIa",
                    se=rep(NA,19),
                    qty=rep(5000,19),
                    qty.rn=c(NA,NA),
                    conc.g1=c(3.24e7,rep(NA,3)),
                    conc.g2=rep(NA,4),
                    expos=rep(1,19),
                    infec=rep(NA,19),
                    sympt=c(rep(1,7),rep(0,12)));
# Dolin et al. 1982: Detection by immune electron microscopy of
the
# Snow Mountain agent of acute viral gastroenteritis.
# Journal of Infectious Diseases 146(2):184-189.
challenge.18 <- list(strain="G2.2: SMV",
                   # se=c(rep(1,2),rep(1,2),rep(1,2),rep(0,1),
                   #      rep(1,3),rep(1,2)),
                   se=c(rep(NA,2),rep(NA,2),rep(NA,2),rep(NA,1),
                        rep(NA,3),rep(NA,2)),

qty=c(rep(1,2),rep(10,2),rep(100,2),rep(500,4),
                         rep(1000,2)),
                   qty.rn=c(NA,NA),
                   conc.g1=rep(NA,4),
                   conc.g2=c(3.17e3,rep(NA,3)),
```

```
                    expos=c(rep(1,2),rep(1,2),rep(1,2),rep(1,4),
                            rep(1,2)),
                    infec=c(rep(0,2),rep(1,2),rep(1,2),rep(0,1),
                            rep(1,3),rep(1,2)),
                    sympt=c(rep(0,2),rep(1,2),rep(1,2),rep(0,1),
                            rep(1,3),rep(1,2)));
# Chapel Hill SMV challenge study
challenge.19 <- list(strain="G2.2: SMV",
                    se=c(rep(0,1),rep(1,4),rep(0,1),rep(1,4),
                         rep(0,1),rep(1,4)),
                    qty=c(rep(1e-2,1),rep(1e-
2,4),rep(1,1),rep(1,4),
                          rep(1e2,1),rep(1e2,4)),
                    qty.rn=c(NA,NA),
                    conc.g1=rep(NA,4),
                    conc.g2=c(3.17e3,rep(NA,3)),
                    expos=c(rep(1,1),rep(1,4),rep(1,1),rep(1,4),
                            rep(1,1),rep(1,4)),
                    infec=c(rep(0,1),rep(0,4),rep(0,1),rep(1,4),
                            rep(1,1),rep(1,4)),
                    sympt=c(rep(0,1),rep(0,4),rep(0,1),rep(0,1),

rep(1,3),rep(1,1),rep(0,1),rep(1,3)));
# Frenck et al (2012) Predicting susceptibility to norovirus
GII.4
# by use of a challenge model involving humans
# Journal of Infectious Diseases 206(9):1386-1393
challenge.20 <- list(strain="G2.4",
                    se=c(rep(0,17),rep(1,23)),
                    qty=rep(1,40),
                    qty.rn=c(NA,NA),
                    conc.g1=rep(NA,4),
                    conc.g2=c(5.0e4,rep(NA,3)),
```

```
                    expos=rep(1,40),
                    infec=c(rep(0,16),rep(1,1),
                            rep(0,7),rep(1,16)),
                    sympt=c(rep(0,16),rep(0,1),
                            rep(0,10),rep(1,13)));
# Bernstein et al (2015) Norovirus Vaccine Against Experimental
Human
# GII.4 Virus Illness: A Challenge Study in Healthy Adults.
# Journal of Infectious Diseases 211:870-878
challenge.21 <- list(strain="G2.4",
                    se=rep(1,48),
                    qty=rep(1,48),
                    qty.rn=c(NA,NA),
                    conc.g1=rep(NA,4),
                    conc.g2=c(4.4e3,rep(NA,3)),
                    expos=rep(1,48),
                    infec=c(rep(1,30),rep(0,18)),
                    sympt=c(rep(1,18),rep(0,30)));


hstnam  <- c("challenge");
# n.challenge <- 21;
# stnum   <- c( 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14,
15, 16, 17, 18,
#              19,20,21);
# expnum  <- c( 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14,
15, 16, 17, 18,
#              19,20,21);
# strains <- c( 1, 1, 1, 1, 1, 1, 1, 1, 1,  1,  1,  1,  1,  1,
1,  1,  1,
#               2, 2, 2, 2);
# hosts   <- c( 1, 1, 1, 1, 1, 1, 1, 1, 1,  1,  1,  1,  1,  1,
1,  1,  1,  1,
#               1, 1, 1);
```

```
# n.dose  <- c(75,33,13,15,49,50,42,59,16,  7, 12, 15, 15, 16,
36,  7, 19, 12,
#              15,40,48);


### All GI.1 Studies ###
# n.challenge <- 17;
# stnum   <- c( 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14,
15, 16, 17);
# expnum  <- c( 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14,
15, 16, 17);
# strains <- c( 1, 1, 1, 1, 1, 1, 1, 1, 1,  1,  1,  1,  1,  1,
1,  1,  1);
# hosts   <- c( 1, 1, 1, 1, 1, 1, 1, 1, 1,  1,  1,  1,  1,  1,
1,  1,  1);
# n.dose  <- c(75,33,13,15,49,50,42,59,16,  7, 12, 15, 15, 16,
36,  7, 19);


### New Studies ###
 n.challenge <- 5;
 stnum   <- c( 1, 2, 3, 4, 5);
 expnum  <- c( 1, 2, 3, 4, 5);
 strains <- c( 1, 1, 1, 1, 1);
 hosts   <- c( 1, 1, 1, 1, 1);
 n.dose  <- c(75,33,13,15,49);


strain <- c(); se <- c(); intk <- c(); intk.rn <- c();
conc <- array(NA,dim=c(max(expnum),2,4));
expos <- c(); infec <- c(); sympt <- c(); exn <- c();
stnam <- c(); obn <- c();
for(k in 1:length(n.dose)){
  strain <- c(strain,rep(strains[k],n.dose[k]));
  stn <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep="")))$str
ain;
  exn <- c(exn,rep(expnum[k],n.dose[k]));
```

```
  qty <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$qty
;

  qrn <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$qty
.rn;

  cg1 <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$con
c.g1;

  cg2 <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$con
c.g2;

  exs <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$exp
os;

  ses <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$se;

  inf <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$inf
ec;

  smp <-
eval(parse(text=paste(hstnam[hosts[k]],".",stnum[k],sep=""")))$sym
pt;

  stnam <- c(stnam,stn);

  intk <- c(intk,qty[1:n.dose[k]]);

  intk.rn <- rbind(intk.rn,as.vector(qrn));

  conc[k,1,] <- as.vector(cg1);

  conc[k,2,] <- as.vector(cg2);

  se <- c(se,ses[1:n.dose[k]]);

  expos <- c(expos,exs[1:n.dose[k]]);

  infec <- c(infec,inf[1:n.dose[k]]);

  sympt <- c(sympt,smp[1:n.dose[k]]);

}

aggr <- rep(0,length(exn));

aggr[exn==1] <- 1;

n.ch <- length(which(exn<=n.challenge));

n.ob <- max(exn)-n.challenge;
```

```
last <- length(exn);
do.ch <- which(exn<=n.challenge);
n.str <- 1; ch.str <- rep(NA,last);
for(k in do.ch){
  for(k.str in 1:n.str){
    if(any(!is.na(conc[exn[k],k.str,]))) ch.str[k] <- k.str;
  }
}
do.ob <- which(exn>n.challenge);
n.conc <- array(NA,dim=c(max(exn),2));
for(k.exp in 1:max(exn)){
  for(k.st in 1:2){
    n.conc[k.exp,k.st] <-
length(which(!is.na(conc[k.exp,k.st,])));
  }
}
sep <- rep(NA,length(exn)); sep[do.ch] <- 2; sep[do.ob] <- 3;

se.init <- rep(NA,last);
for(k in 1:last){
  if(is.na(se[k]) & !is.na(infec[k])) se.init[k] <- infec[k];
  if(is.na(se[k]) & is.na(infec[k])) se.init[k] <- sympt[k];
}

n.hs <- 2; # Se+, Se-

prior.w <- array(c(-12,0,-12,0,
                   1,0.01,1,0.01),dim=c(n.hs,2,2));#/2;
prior.z <- rbind(c(0,1),c(0,1));#/2;
tau.w <- rbind(c(1,0.05),c(0.05,0.05));

prior.rho.a <- c(2,4e3);
```

```
prior.rho.c <- c(4,10);
prior.lambda.c <- c(4,1e3);
prior.se <- c(79,19);

drdata <- list("prior.w"=prior.w,"tau.w"=tau.w,"prior.z"=prior.z,
               "prior.rho.a"=prior.rho.a,
               "prior.se"=prior.se,
               "do.ch"=do.ch,
               "n.str"=n.str,"ch.str"=ch.str,
               "exn"=exn,
               "conc"=conc,
               "n.hs"=n.hs,"se"=se,"aggr"=aggr,"sep"=sep,
               "intk"=intk,"expos"=expos,"infec"=infec,
               "sympt"=sympt);

drinit <- list("se"=se.init);

library(rjags);

ver <- "v9a";

# ".RNG.state" <- c(19900, 14957, 25769)
n.burn <- 1000;
n.iter <- 5000;
n.chains <- 4;
n.post <- 1000;
n.thin <- round(n.iter/n.post);
n.post <- n.iter/n.thin;

tomonitor <- c("a","b","intk","dose","lambda.agg[51,1]","rho.a");

# file names
```

```
file.mod <- paste(ver,".model.","jags",sep="");
file.res <- paste("./output/",ver,".result.","pdf",sep="");
file.txt <- paste("./output/",ver,".result.","txt",sep="");
file.dat <- paste(ver,".data.","r",sep="");
file.pst <- paste("./output/",ver,".post.","rda",sep="");
file.par <- paste("./output/",ver,".mcmc.","rda",sep="");
file.xtr <- paste(ver,".extract.","r",sep="");
file.pgr <- paste(ver,".graph.","r",sep="");
file.dgr <- paste(ver,".dore.","r",sep="");
file.idx <- paste(ver,".idx.","r",sep="");
file.sta <- paste(ver,".stats.","r",sep="");


# loading the data
source(file.dat);


cat("<<< Compile model >>>\n");
# mod.pst <-
jags.model(file=file.mod,data=drdata,n.chains=n.chains,inits=drin
it);
mod.pst <-
jags.model(file=file.mod,data=drdata,n.chains=n.chains);
update(mod.pst,n.burn=n.burn);
cat("<<< MCMC Sampling >>>\n");
mcmc.pst <-
coda.samples(mod.pst,tomonitor,n.iter=n.iter,thin=n.thin);


# initsfunction <- function(chain){
#   stopifnot(chain %in% (1:4)); # max 4 chains allowed...
#   .RNG.seed <- (1:4)[chain];
#   .RNG.name <- c("base::Wichmann-Hill","base::Marsaglia-
Multicarry",
#                  "base::Super-Duper","base::Mersenne-
Twister")[chain];
#   return(list(".RNG.seed"=.RNG.seed,
```

```
#                ".RNG.name"=.RNG.name));
#   inits=drinit;
# }

# cat("<<< Compile model >>>\n");
# cat("<<< MCMC Sampling >>>\n");
# mcmc.pst <- run.jags(model=file.mod,data=drdata,
#                      inits=initsfunction,method="parallel",
#                      burnin=n.burn,thin=n.thin,sample=n.post,
#                      n.chains=n.chains,
#                      monitor=tomonitor);

# cat("<<< Store summary >>>\n");
# sink(file.txt);
# print(summary(mcmc.pst));
# sink();
# cat("<<< Graphing results >>>\n");
# pdf(file.res);
# plot(mcmc.pst,trace=TRUE,density=FALSE);
# plot(mcmc.pst,trace=FALSE,density=TRUE);
# dev.off();
cat("<<< Store posterior >>>\n");
save(mcmc.pst,file=file.pst,ascii=TRUE);
cat("<<< Store parameter estimates >>>\n");
source(file.xtr);
cat("<<< Parameter scattergraphs >>>\n");
source(file.pgr);
cat("<<< Graph dose response relations >>>\n");
source(file.dgr);
cat("<<< Graph infdx >>>\n");
source(file.idx);
cat("<<< Calculate statistics >>>\n");
```

```
source(file.sta);


library(coda);


file.par <- paste("./output/",ver,".mcmc.","rda",sep="");
file.dos <- paste("./output/",ver,".mcmc-dose.","rda",sep="");


mcmc.mat <- as.matrix(as.mcmc.list(mcmc.pst));


extr.var <- function(nam,index){
  var.mc <- c();
  varname <- paste(nam,"[",sep="");
  if(length(index)==1) varname <-
paste(varname,index,"]",sep="");
  if(any(is.na(index))) varname <- nam;
  if(length(index)>1){
    for(k.index in 1:(length(index)-1)){
      varname <- paste(varname,index[k.index],",",sep="");
    }
    varname <- paste(varname,index[length(index)],"]",sep="");
  }
  var.mc <- mcmc.mat[,which(colnames(mcmc.mat)==varname)];
  return(var.mc);
}


a.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
b.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
r.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
eta.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    a.mc[k.hs,k.str,] <- extr.var("a",c(k.hs,k.str,1));
```

```
    b.mc[k.hs,k.str,] <- extr.var("b",c(k.hs,k.str,1));
    r.mc[k.hs,k.str,] <- extr.var("a",c(k.hs,k.str,2));
    eta.mc[k.hs,k.str,] <- extr.var("b",c(k.hs,k.str,2));
  }
}
dose.mc <- array(NA,dim=c(last,n.str,n.chains*n.post));
intk.mc <- array(NA,dim=c(last,n.chains*n.post));
# lambda.mc <- array(NA,dim=c(last,n.str,n.chains*n.post));
# for(k in which(exn==1 | exn==2 | exn==5)){
for(k in 1:last){
  dose.mc[k,1,] <- extr.var("dose",c(k,1));
}
# for(k in which(exn==3 | exn==4 | exn==6 | exn==7)){
#   dose.mc[k,2,] <- extr.var("dose",c(k,2));
# }
# for(k in do.ob){
#   intk.mc[k,] <- extr.var("intk",k);
#   for(k.str in 1:n.str){
#     lambda.mc[k,k.str,] <- extr.var("lambda",c(k,k.str));
#   }
# }
lambda.agg.mc <- extr.var("lambda.agg",c(51,1));
rho.agg.mc <- extr.var("rho.a",NA);
alogser.mc <- lambda.agg.mc/(lambda.agg.mc+rho.agg.mc);
agg.mc <- -alogser.mc/((1-alogser.mc)*log(1-alogser.mc));

# save mcmc values
mc.param <-
list("a.mc"=a.mc,"b.mc"=b.mc,"r.mc"=r.mc,"eta.mc"=eta.mc);
mc.dose <- list("dose.mc"=dose.mc,#"intk.mc"=intk.mc,
                #"lambda.mc"=lambda.mc,
                "agg.mc"=agg.mc);
```

```
save(mc.param,file=file.par,ascii=TRUE);
save(mc.dose,file=file.dos,ascii=TRUE);

library(MASS);
library(Hmisc);
source("hg.r")
source("minticks.r");

# file names
file.dat <- paste(ver,".data.","r",sep="");
file.gra <- function(nam)
return(paste("./output/eps/cont/",nam,".pdf",sep=""));
file.par <- paste("./output/",ver,".mcmc.","rda",sep="");

epsw <- 6;
epsh <- 6;

hs.nam  <- c("Se-","Se+");
str.nam <- c("G1","G2");
# hs.inf  <- c(1,1,2,2);
# str.inf <- c(1,2,1,2);
# hs.ill  <- c(1,1,2,2);
# str.ill <- c(1,2,1,2);
hs.inf  <- c(2);
str.inf <- c(1);
hs.ill  <- c(2);
str.ill <- c(1);

n.plot <- 3000;
choose <- sample((1:(n.chains*n.post)),size=n.plot);

# obtain mcmc sample from file
```

```
load(file.par);
a.mc   <- mc.param$a.mc;
b.mc   <- mc.param$b.mc;
r.mc   <- mc.param$r.mc;
eta.mc <- mc.param$eta.mc;
# par.mc <- list(a.mc,b.mc,r.mc,eta.mc);

w1.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
z1.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
w2.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
z2.mc <- array(NA,dim=c(n.hs,n.str,n.chains*n.post));
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    u1.mc <-
a.mc[k.hs,k.str,]/(a.mc[k.hs,k.str,]+b.mc[k.hs,k.str,]);
    v1.mc <- a.mc[k.hs,k.str,]+b.mc[k.hs,k.str,];
    w1.mc[k.hs,k.str,] <- log(u1.mc/(1-u1.mc));
    z1.mc[k.hs,k.str,] <- log(v1.mc);
    u2.mc <- r.mc[k.hs,k.str,]/
             (r.mc[k.hs,k.str,]+eta.mc[k.hs,k.str,]);
    v2.mc <- r.mc[k.hs,k.str,]+eta.mc[k.hs,k.str,];
    w2.mc[k.hs,k.str,] <- log(u2.mc/(1-u2.mc));
    z2.mc[k.hs,k.str,] <- log(v2.mc);
  }
}

findqlev <- function(dens.est,xvec,yvec,alpha){
  z <- array()
  for (k in 1:(n.chains*n.post)){
    z.x <- which(dens.est$x < xvec[k]);
    z.y <- which(dens.est$y < yvec[k]);
    z[k] <- NA;
```

```
    if(length(z.x) > 0 & length(z.y) > 0) z[k] <-
dens.est$z[max(z.x),max(z.y)];
  }
  clev <- quantile(z,probs=1-alpha,na.rm=TRUE);
  return(clev);
}

mc.cont <- function(x.mc,y.mc,alpha,colour,ltype,dl=FALSE){
  sel <- which(x.mc!=Inf & x.mc!=-Inf & y.mc!=Inf & y.mc!=-Inf);
  dens <- kde2d(x=x.mc[sel],y=y.mc[sel],n=100);
  qlev <- findqlev(dens,x.mc,y.mc,alpha);
  contour(dens,levels=qlev,labels=alpha,frame.plot=FALSE,
          lty=ltype,col=colour,drawlabels=dl,add=TRUE);
}

pos.leg <- function(ax.rng,sz){
 x <- ax.rng[1,1]+sz[1]*(ax.rng[1,2]-ax.rng[1,1]);
 y <- ax.rng[2,1]+sz[2]*(ax.rng[2,2]-ax.rng[2,1]);
 return(c(x,y));
}

infrng <- rbind(c(-10,1),c(-4,4));
illrng <- rbind(c(-20,40),c(-6,5));

col.cnt.inf <- c("gray","gray","black","black");
lty.inf <- c(2,1,2,1);
lab.inf <- c();
for(k in 1:length(str.inf))
  lab.inf <- c(lab.inf,paste(hs.nam[hs.inf[k]],
                             str.nam[str.inf[k]],sep=" "));

setEPS();
```

```
pdf(file.gra("inf-contour"));

par(mar=c(4,4,2,0)+0.1);

plot(x=-1,y=-
1,xlim=infrng[1,],ylim=infrng[2,],col="white",xlab="w",ylab="z");

for(k in 1:length(str.inf)){

  mc.cont(w1.mc[hs.inf[k],str.inf[k],],

          z1.mc[hs.inf[k],str.inf[k],],0.95,

          col=col.cnt.inf[k],ltype=lty.inf[k]);

}

lpos <- pos.leg(infrng,c(0.0,0.27))+c(9.1,-1);

legend(lpos[1],lpos[2],lab.inf,lty=lty.inf,cex=0.75,col=col.cnt.i
nf);

dev.off();


setEPS();

pdf(file.gra("ill-contour-ch"));

par(mar=c(4,4,2,0)+0.1);

plot(x=-1,y=-
1,xlim=illrng[1,],ylim=illrng[2,],col="white",xlab="w",ylab="z");

for(k in 1:length(str.ill)){

  mc.cont(w2.mc[hs.ill[k],str.ill[k],],

          z2.mc[hs.ill[k],str.ill[k],],0.95,

          col=col.cnt.inf[k],ltype=lty.inf[k]);

}

lpos <- pos.leg(illrng,c(0.0,0.27))+c(47.5,-1.4);

legend(lpos[1],lpos[2],lab.inf,lty=lty.inf,cex=0.75,col=col.cnt.i
nf);

dev.off();


library(Hmisc);

source("hg.r")

source("minticks.r");
```

```
# file names
file.dat <- paste(ver,".data.","r",sep="");
file.gra <- function(nam)
return(paste("./output/eps/dr/",nam,".pdf",sep=""));
file.par <- paste("./output/",ver,".mcmc.","rda",sep="");
file.dos <- paste("./output/",ver,".mcmc-dose.","rda",sep="");
file.int <- paste("./output/",ver,".mcmc-intk.","rda",sep="");

epsw <- 6;
epsh <- 4;

hs.nam <- c("Se-","Se+");
str.nam <- c("G1","G2");

n.plot <- 1000;
choose <- sample((1:(n.chains*n.post)),size=n.plot);

# obtain mcmc sample from file
load(file.par);
a.mc   <- mc.param$a.mc;
b.mc   <- mc.param$b.mc;
r.mc   <- mc.param$r.mc;
eta.mc <- mc.param$eta.mc;
load(file.dos);
dose.mc <- mc.dose$dose.mc;
intk.mc <- mc.dose$intk.mc;
lambda.mc <- mc.dose$lambda.mc;

mkdosegroups.ch <- function(n.exp,n.hs,intake,xpos,infc,smpt){
  n.lst <- which(exn==n.exp & se==n.hs);
  intk.lst <- unique(intake[n.lst]);
  dose.grp <- c();
```

```
  for(intk in intk.lst){
    grouped <- which(exn==n.exp & se==n.hs & intake==intk);
    exposed <- sum(xpos[grouped]);
    infectd <- sum(infc[grouped]);
    symptom <- sum(smpt[grouped]);
    dose.grp <- rbind(dose.grp,c(exposed,infectd,symptom));
  }
  return(dose.grp);
}


finddoses.ch <- function(n.exp,n.hs,intake){
  n.lst <- which(exn==n.exp & se==n.hs);
  intk.lst <- unique(intake[n.lst]);
  intk.ind <- c();
  for(intk in intk.lst){
    intk.ind <- c(intk.ind,(which(exn==n.exp & se==n.hs &
intake==intk))[1]);
  }
  return(intk.ind);
}


mkdoses.ch <- function(n.exp,n.hs,intake,n.str){
  dose.ind <- finddoses.ch(n.exp,n.hs,intk); mndose <- c();
  for(k in dose.ind){
    mndose <- c(mndose,mean(dose.mc[k,n.str,]));
  }
  return(mndose);
}


graphdata <-
function(dvec,nvec,kvec,pchoice,colour,connect=FALSE){
  use0 <- which(!is.na(dvec));
```

```
  x <- unique(dvec[use0]);
  n0 <- rep(NA,length(x));
  n1 <- rep(NA,length(x));
  for(k in 1:length(x)){
    use <- which(dvec[use0]==x[k])
    if(length(na.omit(nvec[20]))!=0 &
       length(na.omit(kvec[20]))!=0){
       n0[k] <- sum(na.omit(nvec[20]));
       n1[k] <- sum(na.omit(kvec[20]));
    }
  }
  ord <- order(x);
  x <- log10(x[ord]);
  y <- n1[ord]/n0[ord];
  sz <- 2*sqrt(n0[ord])/2;
  if(connect) lines(x,y);
  if(pchoice > 25) pchoice <- pchoice - 25;
  points(x,y,pch=pchoice,bg=colour,cex=sz);
}


# Graph quantiles of the hypergeometric infection dose response
relation
drawinfdr <- function(alist,blist,doselist,title) {
  nsim <- length(alist)
  ndoses <- length(doselist);
  uu <- rep(NA,nsim);
  qq <- matrix(NA,ndoses,3);

  for(n in 1:ndoses) {
    for(k in 1:nsim) {
      uu[k] <- drinf(alist[k],blist[k],doselist[n]);
    }
```

```
    qq[n,] <- quantile(uu,c(0.025,0.5,0.975),na.rm=TRUE);
  }
  plot(log10(doselist),c(0,rep(1,ndoses-1)),
       ylim=c(0,1),
       main=title,
       ylab="P(Inf)",
       xlab="dose",xaxt="n",
       type="n");
  colo <- c("2.5%","50%","97.5%");
  dimnames(qq) <- list(NULL,colo);
  lin <- 1:ndoses;
  lines(log10(doses),qq[lin,colo[2]],lty=1);
  lines(log10(doses),qq[lin,colo[1]],lty=2);
  lines(log10(doses),qq[lin,colo[3]],lty=2);
  ticks.log(1,n.major=6);
}

# Graph quantiles of the illness dose response relation
drawilldr <- function(alist,blist,rlist,etalist,doselist,title) {
  nsim <- length(alist)
  ndoses <- length(doselist);
  uu <- rep(NA,nsim);
  qq <- matrix(NA,ndoses,3);

  for(n in 1:ndoses) {
    for(k in 1:nsim) {
      uu[k] <- drinf(alist[k],blist[k],doselist[n])*
        drill(rlist[k],etalist[k],doselist[n])
    }
    qq[n,] <- quantile(uu,c(0.025,0.5,0.975),na.rm=TRUE);
  }
  plot(log10(doselist),c(0,rep(1,ndoses-1)),
```

```
        ylim=c(0,1),
        main=title,
        ylab="P(Ill)",
        xlab="dose",xaxt="n",
        type="n");
  colo <- c("2.5%","50%","97.5%");
  dimnames(qq) <- list(NULL,colo);
  lin <- 1:ndoses;
  lines(log10(doses),qq[lin,colo[2]],lty=1);
  lines(log10(doses),qq[lin,colo[1]],lty=2);
  lines(log10(doses),qq[lin,colo[3]],lty=2);
  ticks.log(1,n.major=6);
}

# Graph quantiles of the single hit probability pm
drawpm <- function(alist,blist,ulist,ymax,title) {
  nsim <- length(alist)
  nu <- length(ulist);
  uu <- rep(NA,nsim);
  qq <- matrix(NA,nu,3);

  for(n in 1:nu) {
    for(k in 1:nsim) {
      uu[k] <- pmdist(alist[k],blist[k],ulist[n])
    }
    qq[n,] <- quantile(uu,c(0.05,0.5,0.95),na.rm=TRUE);
  }
  colo <- c("2.5%","50%","97.5%");
  dimnames(qq) <- list(NULL,colo);
  lin <- 1:nu;
  ymx <- max(qq[lin,colo[3]]);
  if(ymx!=Inf) ymax <- ymx;
```

```
  plot(ulist,c(0,rep(1,nu-1)),
       ylim=c(0,ymax),
       main=title,
       ylab="f(pm)",
       xlab="pm",xaxt="n",
       type="n");
  lines(ulist,qq[lin,colo[2]],lty=1);
  lines(ulist,qq[lin,colo[1]],lty=2);
  lines(ulist,qq[lin,colo[3]],lty=2);
  ticks.logit(1);
}

# Define list of doses and pms for graphs
doses <- 10^seq(-2,13,0.25);
pms <- seq(-12,10,0.25);

# Make graphs and write to pdf file
setEPS();

dta.inf <- array(NA,dim=c(2,2,5));
# dta.inf[1,1,] <- c( 1, 2, 5,NA,NA); # Se- G1 Ch
# dta.inf[1,2,] <- c( 4,NA,NA,NA,NA); # Se- G2 Ch
# dta.inf[2,1,] <- c( 1, 2, 5,NA,NA); # Se+ G1 Ch
# dta.inf[2,2,] <- c( 4, 6, 7,NA,NA); # Se+ G2 Ch
dta.ill <- array(NA,dim=c(2,2,5));
# dta.ill[1,1,] <- c( 1, 2, 5,NA,NA); # Se- G1 Ch
# dta.ill[1,2,] <- c( 4,NA,NA,NA,NA); # Se- G2 Ch
# dta.ill[2,1,] <- c( 1, 2, 5,NA,NA); # Se+ G1 Ch
# dta.ill[2,2,] <- c( 4, 6, 7,NA,NA); # Se+ G2 Ch
dta.inf[2,1,] <- c(1,2,3,4,5); # Se+ G1 Ch
dta.ill[2,1,] <- c(1,2,3,4,5); # Se+ G1 Ch
```

```
# for(k.hs in 1:n.hs){
#   for(k.str in 1:n.str){
for(k.hs in 2:2){
  for(k.str in 1:1){
    cat(hs.nam[k.hs],str.nam[k.str]," ");
    grnam <- paste(hs.nam[k.hs]," ",str.nam[k.str],sep="");
    fnam <- paste("drinf-",k.hs,"-",k.str,sep="");
    pdf(file.gra(fnam));
    par(mar=c(4,4,2,0)+0.1);

drawinfdr(a.mc[k.hs,k.str,choose],b.mc[k.hs,k.str,choose],doses,g
rnam);
    for(k.exp in
dta.inf[k.hs,k.str,!is.na(dta.inf[k.hs,k.str,])]){
      data.grp <- cbind(mkdoses.ch(k.exp,k.hs-1,intk,k.str),
                        mkdosegroups.ch(k.exp,k.hs-
1,intk,expos,infec,sympt));
      graphdata(data.grp[,1],data.grp[,2],data.grp[,3],
                pchoice=20+k.exp,colour="white",connect=TRUE);
    }
    dev.off();
  }
}
cat("\n");
# for(k.hs in 1:n.hs){
#   for(k.str in 1:n.str){
for(k.hs in 2:2){
  for(k.str in 1:1){
    cat(hs.nam[k.hs],str.nam[k.str]," ");
    grnam <- paste(hs.nam[k.hs]," ",str.nam[k.str]," ",sep="");
    fnam <- paste("drill-",k.hs,"-",k.str,sep="");
    pdf(file.gra(fnam));
    par(mar=c(4,4,2,0)+0.1);
```

```
    drawilldr(a.mc[k.hs,k.str,choose],b.mc[k.hs,k.str,choose],
              r.mc[k.hs,k.str,choose],
              eta.mc[k.hs,k.str,choose],doses,grnam);
    for(k.exp in
dta.ill[k.hs,k.str,!is.na(dta.ill[k.hs,k.str,])]){
      data.grp <- cbind(mkdoses.ch(k.exp,k.hs-1,intk,k.str),
                        mkdosegroups.ch(k.exp,k.hs-
1,intk,expos,infec,sympt));
      graphdata(data.grp[,1],data.grp[,2],data.grp[,4],
                pchoice=20+k.exp,colour="white",connect=TRUE);
    }
    dev.off();
  }
}
cat("\n");


# fnam <- paste("pminf-",str,"-",hst,sep="");
# postscript(file.gra(fnam),width=epsw,height=epsh);
# par(mar=c(4,4,2,0)+0.1);
#
drawpm(a.mc[k.hs,k.str,choose],b.mc[k.hs,k.str,choose],pms,0.025,
grnam);
# dev.off();


library(MASS);
library(Hmisc);
source("hg.r")
source("minticks.r");


# file names
file.dat <- paste(ver,".data.","r",sep="");
file.gra <- function(nam)
return(paste("./output/eps/idx/",nam,".eps",sep=""));
file.par <- paste("./output/",ver,".mcmc.","rda",sep="");
```

```
epsw <- 5;
epsh <- 5;

str.nam <- c("G1","G2");
hs.nam <- c("Se-","Se+");

n.plot <- 1000;
choose <- sample((1:(n.chains*n.post)),size=n.plot);

# obtain mcmc sample from file
load(file.par);
a.mc   <- mc.param$a.mc;
b.mc   <- mc.param$b.mc;
r.mc   <- mc.param$r.mc;
eta.mc <- mc.param$eta.mc;

infd50 <- array(NA,dim=c(n.hs,n.str,length(choose)));
illd50 <- array(NA,dim=c(n.hs,n.str,length(choose)));
infd25 <- array(NA,dim=c(n.hs,n.str,length(choose)));
illd25 <- array(NA,dim=c(n.hs,n.str,length(choose)));
infd01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
illd01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
# for(k.hs in 1:n.hs){
#   for(k.str in 1:n.str){
for(k.hs in 2:2){
  for(k.str in 1:1){
    cat(hs.nam[k.hs], str.nam[k.str]," ");
    for(k.iter in 1:length(choose)){
      infd25[k.hs,k.str,k.iter] <- findinfdq(0.25,

a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]]);
```

```
      infd01[k.hs,k.str,k.iter] <- findinfdq(0.01,
a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]]);
      if(k.hs==2){ # secretor positve
        infd50[k.hs,k.str,k.iter] <- findinfdq(0.50,
a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]]);
      }
    }
    for(k.iter in 1:length(choose)){
      illd25[k.hs,k.str,k.iter] <- findilldq(0.25,
a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]],
        r.mc[k.hs,k.str,choose[k.iter]],
        eta.mc[k.hs,k.str,choose[k.iter]]);
      illd01[k.hs,k.str,k.iter] <- findilldq(0.01,
a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]],
        r.mc[k.hs,k.str,choose[k.iter]],
        eta.mc[k.hs,k.str,choose[k.iter]]);
      if(k.hs==2){ # secretor positive
        illd50[k.hs,k.str,k.iter] <- findilldq(0.50,
a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]],
          r.mc[k.hs,k.str,choose[k.iter]],
          eta.mc[k.hs,k.str,choose[k.iter]]);
      }
    }
  }
}
cat("\n");

setEPS();
postscript(file.gra("id50+"),width=4*epsw/3,height=epsh);
```

```
par(mar=c(4,4,2,0)+0.1);
# boxplot(log10(cbind(infd50[2,1,],illd50[2,1,],
#                     infd50[2,2,],illd50[2,2,])),ylim=c(1,9),
#         outline=FALSE,yaxt="n",
#         names=c("Se+ G1\ninf","Se+ G1\nill",
#                 "Se+ G2\ninf","Se+ G2\nill"));
boxplot(log10(cbind(infd50[2,1,],illd50[2,1,])),ylim=c(1,9),
        outline=FALSE,yaxt="n",
        names=c("Se+ G1\ninf","Se+ G1\nill"));
ticks.log(2);
dev.off();

postscript(file.gra("id01+"),width=4*epsw/3,height=epsh);
par(mar=c(4,4,2,0)+0.1);
# boxplot(log10(cbind(infd01[2,1,],illd01[2,1,],
#                     infd01[2,2,],illd01[2,2,])),ylim=c(-2,2),
#         outline=FALSE,yaxt="n",
#         names=c("Se+ G1\ninf","Se+ G1\nill",
#                 "Se+ G2\ninf","Se+ G2\nill"));
boxplot(log10(cbind(infd01[2,1,],illd01[2,1,])),ylim=c(-2,2),
        outline=FALSE,yaxt="n",
        names=c("Se+ G1\ninf","Se+ G1\nill"));
ticks.log(2);
dev.off();

# postscript(file.gra("id01-"),width=4*epsw/3,height=epsh);
# par(mar=c(4,4,2,0)+0.1);
# boxplot(log10(cbind(infd01[1,1,],illd01[1,1,],
#                     infd01[1,2,],illd01[1,2,])),ylim=c(-1,10),
#         outline=FALSE,yaxt="n",
#         names=c("Se- G1\ninf","Se- G1\nill",
#                 "Se- G2\ninf","Se- G2\nill"));
```

```
# ticks.log(2,n.major=5);
# dev.off();

dlist <- list("infd50"=infd50[2,1,],"illd50"=illd50[2,1,],
              "infd25"=infd50[2,1,],"illd25"=illd50[2,1,],
              "infd01"=infd50[2,1,],"illd01"=illd50[2,1,]);
save(dlist,file="id-inf-ill-new.rda",ascii=TRUE);
# rerun with new studies only and change filename, e.g. to "id-
inf-ill-new.rda"!

library(MASS);
library(Hmisc);
source("hg.r")
source("minticks.r");

recalc <- TRUE;

# file names
file.dat <- paste(ver,".data.","r",sep="");
file.sta <- function(nam)
return(paste("./output/stats/",nam,".csv",sep=""));
file.par <- paste("./output/",ver,".mcmc.","rda",sep="");

str.nam <- c("G1","G2");
hs.nam <- c("Se-","Se+");

n.plot <- 1000;
choose <- sample((1:(n.chains*n.post)),size=n.plot);

if(recalc){
  # obtain mcmc sample from file
  load(file.par);
```

```
  a.mc   <- mc.param$a.mc;
  b.mc   <- mc.param$b.mc;
  r.mc   <- mc.param$r.mc;
  eta.mc <- mc.param$eta.mc;
  w.mc <- array(NA,dim=c(n.hs,n.str,2,n.chains*n.post));
  z.mc <- array(NA,dim=c(n.hs,n.str,2,n.chains*n.post));
  for(k.hs in 1:n.hs){
    for(k.str in 1:n.str){
      u.mc <-
a.mc[k.hs,k.str,]/(a.mc[k.hs,k.str,]+b.mc[k.hs,k.str,]);
      v.mc <- a.mc[k.hs,k.str,]+b.mc[k.hs,k.str,];
      w.mc[k.hs,k.str,1,] <- log(u.mc/(1-u.mc));
      z.mc[k.hs,k.str,1,] <- log(v.mc);
      u.mc <- r.mc[k.hs,k.str,]/
        (r.mc[k.hs,k.str,]+eta.mc[k.hs,k.str,]);
      v.mc <- r.mc[k.hs,k.str,]+eta.mc[k.hs,k.str,];
      w.mc[k.hs,k.str,2,] <- log(u.mc/(1-u.mc));
      z.mc[k.hs,k.str,2,] <- log(v.mc);
    }
  }

  infd25 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  illd25 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  infd01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  illd01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  pinf01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  pill01 <- array(NA,dim=c(n.hs,n.str,length(choose)));
  a.par  <- array(NA,dim=c(n.hs,n.str,length(choose)));
  b.par  <- array(NA,dim=c(n.hs,n.str,length(choose)));
  r.par  <- array(NA,dim=c(n.hs,n.str,length(choose)));
  eta.par  <- array(NA,dim=c(n.hs,n.str,length(choose)));
  w.par  <- array(NA,dim=c(n.hs,n.str,2,length(choose)));
```

```
  z.par  <- array(NA,dim=c(n.hs,n.str,2,length(choose)));
  for(k.hs in 1:n.hs){
    for(k.str in 1:n.str){
      cat(hs.nam[k.hs], str.nam[k.str], " ");
      for(k.iter in 1:length(choose)){
        infd25[k.hs,k.str,k.iter] <- findinfdq(0.25,

a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]]);
        infd01[k.hs,k.str,k.iter] <- findinfdq(0.01,

a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]]);
        pinf01[k.hs,k.str,k.iter] <-
          drinf(a.mc[k.hs,k.str,choose[k.iter]],
                b.mc[k.hs,k.str,choose[k.iter]],1.0);
        a.par[k.hs,k.str,k.iter] <-
a.mc[k.hs,k.str,choose[k.iter]];
        b.par[k.hs,k.str,k.iter] <-
b.mc[k.hs,k.str,choose[k.iter]];
        illd01[k.hs,k.str,k.iter] <- findilldq(0.01,

a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]],
          r.mc[k.hs,k.str,choose[k.iter]],
          eta.mc[k.hs,k.str,choose[k.iter]]);
        illd25[k.hs,k.str,k.iter] <- findilldq(0.25,

a.mc[k.hs,k.str,choose[k.iter]],b.mc[k.hs,k.str,choose[k.iter]],
          r.mc[k.hs,k.str,choose[k.iter]],
          eta.mc[k.hs,k.str,choose[k.iter]]);
        pill01[k.hs,k.str,k.iter] <- pinf01[k.hs,k.str,k.iter] *
          drill(r.mc[k.hs,k.str,choose[k.iter]],
                eta.mc[k.hs,k.str,choose[k.iter]],1.0);
        r.par[k.hs,k.str,k.iter] <-
r.mc[k.hs,k.str,choose[k.iter]];
        eta.par[k.hs,k.str,k.iter] <-
```

```
          eta.mc[k.hs,k.str,choose[k.iter]];
        w.par[k.hs,k.str,,k.iter] <-
w.mc[k.hs,k.str,,choose[k.iter]];
        z.par[k.hs,k.str,,k.iter] <-
z.mc[k.hs,k.str,,choose[k.iter]];
      }
    }
  }
}
cat("\n");


calcstats <- function(x){
  x.mn <- mean(x,na.rm=TRUE);
  x.qn <- quantile(x,c(0.5,0.025,0.975),na.rm=TRUE);
  return(c("mean"=x.mn,x.qn));
}


idx.stats <- c();
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    idx.stats <-
rbind(idx.stats,c("InfD25",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(infd25[k.hs,k.str,]),digits=6)));
    idx.stats <-
rbind(idx.stats,c("InfD01",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(infd01[k.hs,k.str,]),digits=6)));
    idx.stats <-
rbind(idx.stats,c("IllD25",hs.nam[k.hs],str.nam[k.str],
        signif(calcstats(illd25[k.hs,k.str,]),digits=6)));
    idx.stats <-
rbind(idx.stats,c("IllD01",hs.nam[k.hs],str.nam[k.str],
        signif(calcstats(illd01[k.hs,k.str,]),digits=6)));
  }
}
```

```
write.table(idx.stats,
            file=file.sta("idx"),row.names=FALSE,
            col.names=c("IdX","Se stat","GG",
                        "mean","P50","P2.5","P97.5"),
            sep=",");


px.stats <- c();
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    px.stats <-
rbind(px.stats,c("Pinf(1)",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(pinf01[k.hs,k.str,]),digits=6)));
    px.stats <-
rbind(px.stats,c("Pill(1)",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(pill01[k.hs,k.str,]),digits=6)));
  }
}


write.table(px.stats,
            file=file.sta("px"),row.names=FALSE,
            col.names=c("PX","Se stat","GG",
                        "mean","P50","P2.5","P97.5"),
            sep=",");


par.stats <- c();
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    par.stats <-
rbind(par.stats,c("a",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(a.par[k.hs,k.str,]),digits=6)));
    par.stats <-
rbind(par.stats,c("b",hs.nam[k.hs],str.nam[k.str],
```

```
      signif(calcstats(b.par[k.hs,k.str,]),digits=6)));
    par.stats <-
rbind(par.stats,c("r",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(r.par[k.hs,k.str,]),digits=6)));
    par.stats <-
rbind(par.stats,c("eta",hs.nam[k.hs],str.nam[k.str],
      signif(calcstats(eta.par[k.hs,k.str,]),digits=6)));
  }
}


write.table(par.stats,
            file=file.sta("par"),row.names=FALSE,
            col.names=c("par","Se stat","GG",
                        "mean","P50","P2.5","P97.5"),
            sep=",");


wz.stats <- c();
for(k.hs in 1:n.hs){
  for(k.str in 1:n.str){
    mn <- c(mean(w.par[k.hs,k.str,1,],na.rm=TRUE),
            mean(z.par[k.hs,k.str,1,],na.rm=TRUE));
    cv <- c(var(w.par[k.hs,k.str,1,]),
            var(w.par[k.hs,k.str,1,],z.par[k.hs,k.str,1,]),
            var(z.par[k.hs,k.str,1,]));
    wz.stats <- rbind(wz.stats,
            c("Inf",hs.nam[k.hs],str.nam[k.str],
              signif(mn,digits=6),signif(cv,digits=6)));
    mn <- c(mean(w.par[k.hs,k.str,2,],na.rm=TRUE),
            mean(z.par[k.hs,k.str,2,],na.rm=TRUE));
    cv <- c(var(w.par[k.hs,k.str,2,]),
            var(w.par[k.hs,k.str,2,],z.par[k.hs,k.str,2,]),
            var(z.par[k.hs,k.str,2,]));
```

```
    wz.stats <- rbind(wz.stats,
            c(paste("Ill ",sep=""),
              hs.nam[k.hs],str.nam[k.str],
              signif(mn,digits=6),signif(cv,digits=6)));
  }
}

write.table(wz.stats,
            file=file.sta("wz"),row.names=FALSE,
            col.names=c("endpoint","Se
stat","GG","mean(w)","mean(z)",
                        "var(w)","cov(w,z)","var(z)"),
            sep=",");

load("id-inf-ill-all.rda");
infd50.all <- dlist$infd50; illd50.all <- dlist$illd50;
infd25.all <- dlist$infd25; illd25.all <- dlist$illd25;
infd01.all <- dlist$infd01; illd01.all <- dlist$illd01;

load("id-inf-ill-new.rda"); # you also need to make this one!
infd50.new <- dlist$infd50; illd50.new <- dlist$illd50;
infd25.new <- dlist$infd25; illd25.new <- dlist$illd25;
infd01.new <- dlist$infd01; illd01.new <- dlist$illd01;

### Comparing Infectivity of All vs. New studies ###
# 50% #
infd50.diff <- infd50.all - infd50.new;
hist(infd50.diff) # show their distribution

length(which(infd50.diff > 0)); # how many are > 0?
length(infd50.diff);            # how many are there in total?
```

```
length(which(infd50.diff > 0))/length(infd50.diff) # fraction > 0

# 1% #
infd01.diff <- infd01.all - infd01.new;
hist(infd01.diff) # show their distribution

length(which(infd01.diff > 0)); # how many are > 0?
length(infd01.diff);            # how many are there in total?

length(which(infd01.diff > 0))/length(infd01.diff) # fraction > 0

### Comparing Illness of All vs. New studies ###
# 50% #
illd50.diff <- illd50.all - illd50.new;
hist(illd50.diff) # show their distribution

length(which(illd50.diff > 0)); # how many are > 0?
length(illd50.diff);            # how many are there in total?

length(which(illd50.diff > 0))/length(illd50.diff) # fraction > 0

# 1% #
illd01.diff <- illd01.all - illd25.new;
hist(illd01.diff) # show their distribution

length(which(illd01.diff > 0)); # how many are > 0?
length(illd01.diff);            # how many are there in total?

length(which(illd01.diff > 0))/length(illd01.diff) # fraction > 0
```