

**Title:** Viral Contamination of Household Wells Near Disposal Sites for Human Excreta

**Project I.D.:** DNR Project #144

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**Background/Need:** Public health officials suspect that contaminated groundwater may be responsible for many cases of endemic enteric disease that are too sporadic to easily identify the infection source. Enteric viruses are the most likely human pathogens to contaminate groundwater. Their extremely small size (around 30 nm) and slow die-off rates allow them to infiltrate and survive in soils, eventually reaching aquifers.

The question confronting public health and regulatory agencies 10 years ago was, do viruses survive and travel through soils? We now know that they do. The question now is, what is the incidence of viral contamination in groundwater? Recent studies monitoring ground water for enteric viruses have focused on municipal wells, whereas the incidence of viruses in private household wells is unknown. However, household wells may be more susceptible to viral contamination than municipal wells because they are shallower, may be less carefully maintained, and can be located in close proximity to areas with high loading rates of human excreta. For regions of the US that rely heavily on ground water, like the northern Midwest, data on virus incidence in private wells would complement the municipal well data to more fully characterize the extent and conditions of groundwater contamination with enteric viruses.

**Objectives:** The objective of this study was to estimate the occurrence of human enteric viruses in private household wells located near sites with high loading rates of human excreta, specifically septage landspreading sites and subdivisions with high densities of septic systems. The rationale for this approach was that if wells near sources of enteric viruses were not contaminated, then other wells in regions with lower excreta loading rates would not likely be contaminated. Secondary objectives included comparing the occurrence of enteric viruses among wells in different hydrogeologic settings, and assessing the predictive value of indicators of water sanitary quality for virus contamination.

**Methods:** Wells were divided into two sets of 25, one set sampled for one year and a new set sampled the second year. Each well was sampled four times, once each season. All wells were privately owned and served a single household. Wells were selected based on their proximity to land-spreading sites of human septage or rural subdivisions with high densities of conventional septic systems. The rationale for this selection scheme was that viruses only would be detected if there is a fecal source and that these wells then would likely represent the upper limit of virus contamination. Viruses were concentrated using a 1 MDS cartridge filter, detected by reverse-transcriptase polymerase chain reaction (RT-PCR), and confirmed by Southern hybridization. In addition, all samples were tested for enteroviruses by cell culture. Virus groups investigated included enteroviruses, rotavirus, hepatitis A, and the human caliciviruses (genogroups 1 and 2). Companion water samples

were collected for water quality indicators including total coliforms, *E.coli*, fecal enterococci, male-specific coliphages, nitrate, and chloride.

**Results and Discussion:**

Among the fifty wells, four (8%) were positive for viruses. Three wells were positive for hepatitis A and the fourth well was positive for both rotavirus and G1 calicivirus in one sample and an enterovirus in another sample. Contamination was transient, as none of the wells was virus-positive for two sequential samples. Enteric virus incidence among samples was 3% (5/194). None of the samples were positive for enteroviruses by cell culture. RT-PCR inhibition occurred in 8% (16/194) of the ground water samples, primarily those collected during the winter. Male-specific coliphages were found in two samples, however, this indicator and the others were not statistically associated with virus occurrence on either a per-sample or per-well basis. The sensitivity and positive predictive value of the indicators were generally low, except for chloride.

**Conclusions/ Implications:**

In the United States it is estimated there are 267 million episodes of acute diarrhea each year (Glass et al 2001). The majority of diarrheal illnesses are endemic (i.e. non-outbreak). How many of these are attributable to drinking water is unknown, let alone the fraction attributable to drinking from contaminated household wells. The relative importance of household wells as a disease transmission route can be gauged from the potential number of people exposed. Fifteen million households in the US use a private well as their primary drinking water source (US Census 1990). Assuming that the 8% virus contamination rate reported in the present study is a reasonable estimate of the national rate, then 1.2 million households are exposed to enteric viruses through their private wells. However, the generalizability of the 8% rate is uncertain. It may be underestimated given that the sanitary quality of Wisconsin groundwater is relatively high compared to other Midwest states. It may be overestimated because the wells in this study were located near fecal sources. What is certain is that some household wells are contaminated with human enteric viruses, presenting a risk for disease transmission that should be investigated further.

**Publications:**

Borchardt, M.A., P.D Bertz, S.K. Spencer, and D.A. Battigelli. In Review. Incidence of enteric viruses in groundwater from household wells in Wisconsin. Appl. Environ. Microbiology.

Borchardt, M.A., P-H Chyou, E.O. DeVries, and E.A. Belongia. In Review. Septic systems and groundwater: Association with infectious diarrhea in a defined population of children. Am. J. Epidemiol.

**Key Words:**

Viruses, private wells, RT-PCR, septic systems, landspreading, microbial indicators

**Funding:**

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**Final Report:**

A final report containing more detailed information on this project is available for loan from Wisconsin's Water Library, University of Wisconsin - Madison, 1975 Willow Drive, Madison, Wisconsin 53706 (608) 262-3069.