ASSESSING GROUNDWATER QUALITY IN KEWAUNEE COUNTY, WISCONSIN

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(Expertise in constructing and installing auto-samplers for collection of pathogen samples)

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(His office will provide support for objective 5)
**Background:**

*Existing Water Quality*

The fractured Silurian dolomite aquifer is an important, but vulnerable, source of drinking water in northeast Wisconsin (Sherrill, 1978; Bradbury and Muldoon, 1992; Muldoon and Bradbury, 2010). Existing data from Kewaunee County indicate contamination by nitrate-N and coliform bacteria. The UW-Stevens Point Center for Watershed Science database of private well water quality indicates that 12% of 857 samples for nitrate-N exceed the drinking water standard of 10 mg/L. Twenty percent of 720 samples tested positive for coliform bacteria; 16% of the 142 positive coliform bacteria water samples also tested positive for *E. coli*.

Using only *E. coli* as an indication of human or animal waste likely underestimates the number of wells that are contaminated with fecal types of bacteria or other pathogens (Atherholt et al. 2003; Braatz, 2004). While a positive *E. coli* test confirms human or animal waste source contamination, the absence of *E. coli* does not guarantee that a well is free of pathogenic microorganisms. The lack of cost-effective diagnostic tests has limited the ability of homeowners to understand the prevalence of pathogenic groundwater contamination in northeastern Wisconsin wells.

Figure 1 summarizes the nitrate-N and bacteria data for Kewaunee County that have been reported to the UW-Stevens Point Center for Watershed Science. Exceedances of groundwater standards are not randomly distributed but rather appear to correlate with areas where thin soils overlie the fractured dolomite aquifer.

![Figure 1. Existing water-quality data for Kewaunee County (Bonness and Masarik, 2014)](image-url)
Source of Contamination

As resource managers try to address these water-quality problems, there is no consensus as to whether the main source of contamination is human or bovine wastewater. In Kewaunee County, dairy farming and associated crop production comprise the primary land use and manure is commonly applied to crop land prior to spring planting and again in fall after crops have been harvested. In addition, crop land receives commercial fertilizers, septic wastes, as well as industrial and municipal wastes. All of these land-applied materials are long-term, diffuse sources that can contribute to nitrate contamination. Fecal wastes from humans and dairy cattle can be diffuse, long-term sources of enteric pathogens. Prior to developing this proposal, we conducted a pilot project to assess whether sampling private wells for viruses could be an effective method of assessing sources of wastewater impacts. Samples from 10 wells in northern Kewaunee County, collected in May 2014, revealed that seven of the ten wells were positive for fecal contamination. Two wells contained human-specific viruses, one well contained bovine-specific viruses, one well contained both virus types, and one well was positive for bovine Bacteroides. Salmonella species and Campylobacter jejuni were identified in four wells and one well, respectively, which is a human health concern.

Pathogens in Groundwater

Contamination of North American groundwater by human enteric pathogens is well established in the scientific literature (Hynds et al. 2014) resulting in notable disease outbreaks (Wallender et al. 2014) and less obvious levels of illness that are sporadic but, nonetheless, measurable (Borchardt et al 2012). Wallender et al. (2014) recently reported that of the 248 documented outbreaks during 1971 to 2008 related to untreated groundwater, 26% happened in hydrogeological settings similar to northeastern Wisconsin. In Door County, Wisconsin in 2007, 229 people became severely ill after drinking norovirus-contaminated well water at a new restaurant. Dye trace tests showed rapid transport to the restaurant’s well from the septic tanks, where a broken connecting pipe was later discovered, and from the septic leach field located upgradient from the well (Borchardt et al. 2011).

Nitrate in Groundwater

Nitrogen is a widespread and vexing contaminant to ground and surface water, affecting the drinking water resources and the biotic integrity of many of the nation’s water bodies (USEPA, 2009; USGS 2010). Background or natural levels of nitrate-nitrogen in groundwater are generally less than 1 mg/L. Concentrations above 1 mg/L indicate influence by one or more of the following sources: nitrogen fertilizers, manure or other bio-solids (both application to land-surface or leakage from storage), or septic system drainfields. Nitrate-nitrogen concentrations above the drinking water standard of 10 mg/L should not be consumed by infants or women who are pregnant or...
expecting to become pregnant, all other persons are encouraged to avoid long-term consumption of water greater than 10 mg/L (WI DNR, 2014).

Recharge to the Dolomite Aquifer:
Previous work in NE Wisconsin has demonstrated that recharge to the aquifer can be exceedingly rapid (e.g., Bradbury and others, 2002; Muldoon and Bradbury, 2010). Figure 2 illustrates variation in fluid temperature, electrical conductivity, and water levels for a shallow monitoring well in Kewaunee County. Recharge events, indicated by sharp rises in water-level, are both rapid and episodic throughout the year. Changes in fluid conductivity in response to recharge indicate the complexities of the recharge process. The lower graph indicates a rain event in early December that led to no groundwater recharge as the ground was frozen. But then a second rain event in late December, when the temperature was above freezing for several days, produced a sharp rise in conductivity (as vadose water drains) followed by a drop in conductivity as low-conductivity recharge water enters the aquifer. While these data illuminate the timing and rapidity of recharge events to the dolomite aquifer, we have very limited understanding on the delivery and transport of enteric pathogens within this aquifer. The fundamental question is when do the enteric pathogens enter the aquifer? Is it during the water-level rise? Is it when conductivity is rising? Or when conductivity is decreasing? Is it a specified time after the rain event? If we understood pathogen breakthrough, in relation to these indicators of groundwater recharge, we could design better sampling protocols.

Proposed work:
The proposed project seeks to quantify the extent of groundwater contamination by nitrate-N, sanitary quality indicator bacteria, and enteric pathogens in Kewaunee County.

Figure 2. Top: Variation in water level, fluid temperature and electrical conductivity as indicators of recharge in well KW183 along with the precipitation record (from Muldoon and Bradbury, 2010). The top graph indicates that there were five recharge events during the study period. The lower graph illustrates the details of the recharge event in December 2007.
Objectives:

1) Design a randomized, synoptic sampling plan that evaluates groundwater quality throughout Kewaunee County. The randomized sample will be stratified by depth to bedrock. Collect and analyze samples from the randomized sampling frame for nitrate and indicator bacteria.

2) Sample a subset of wells from the randomized and stratified sampling frame on a bi-weekly basis (i.e., once every two weeks) to assess seasonal variation in groundwater quality.

3) Sample a subset of wells once per season (four times per year) for viruses and fecal markers capable of distinguishing septic versus bovine sources of contamination.

4) Install an automated sampling system on one or more wells in order to determine the timing of peak transport for viruses and indicator bacteria.

5) Perform statistical analysis of samples collected from this study and of existing water-quality data from Kewaunee County to assess whether these data can shed light on spatial and temporal patterns of contamination.

Project Approach

Objective 1: Design a randomized synoptic sampling plan to assess extent of groundwater contamination in Kewaunee County. The randomized sampling will be stratified by depth to bedrock.

We will accomplish this objective by obtaining the list of property parcels in Kewaunee County and using a threshold property value to eliminate parcels that do not contain houses. This effort is focused on private drinking water supply wells and thus we will exclude parcels within the areas that have a municipal water supply system – Algoma, Kewaunee and Luxemburg. Properties connected to municipal water can be identified from utility billing addresses, which we will request from the three utilities. From the parcels identified as owning a private well, we will randomly select 563 wells and we will send well owners a letter soliciting their study participation. From a previous private well study using a similar approach (Borchardt et al 2003) we anticipate an 80% participation rate resulting in a sampling frame of 450 private domestic wells. The total number of wells in Kewaunee County is estimated to be ~3900. The Wisconsin Geological and Natural History Survey (WGNHS) has 2029 well records for the period 1936-1989. The online, DNR database indicates that an additional 1881 wells were drilled since 1989. Presumably some of the older wells have been abandoned and well constructions reports (WCRs) might not have been submitted for every well. The proposed sample size represents approximately 11% of the wells in Kewaunee County.

The random sampling frame will be stratified by depth to bedrock based on existing maps (Figure 1) and well construction reports when available. The reason for using a stratified random sampling design is to provide a more precise estimate of the county-wide well contamination rate and to ensure that wells with different depths to
bedrock are equally represented.

Sample collection and analysis will be conducted by staff of the Environmental Research and Innovation Center (ERIC lab) at UW-Oshkosh. Homeowners will be sent sample bottles and instructions on proper sample collection techniques and samples will be dropped off by homeowners on specified dates at designated sample drop-off sites. We anticipate at least 3 drop-off sites: Algoma, Luxemburg, and Kewaunee. This approach has been used successfully by ERIC staff for a study of groundwater quality in Door County.

Sampling of all randomly selected wells will happen on two dates, one during a period of recharge and one during a non-recharge period. Previous work (Muldoon and Bradbury, 2010) suggests that recharge events tend to dilute concentrations of conservative contaminants (such as nitrate-N and chloride) and increase detections for bacteria and viruses. A late October event would correspond to a period of manure application and, depending on precipitation, this could also be a period of groundwater recharge. A second sampling event, either during a period of frozen ground, or during the growing season would correspond to non-recharge conditions. Spatially the proposed sampling design is random, but temporally the design is weighted towards the times when groundwater is most vulnerable to contamination by the various parameters of interest. **In other words, we designed the sampling to give information on the proportion of contaminated wells and not to determine the proportion of contaminated samples.** Even with weighting the sampling times to when groundwater is most vulnerable, the proportion of wells that are contaminated will likely be underestimated because of the rapid, transient nature of contamination events in this aquifer.

Samples will be analyzed for nitrate-N, total coliforms, and *E. coli*. Please note that while the proposal RFP requests information on fecal coliforms, this is not a conventional test for groundwater microbial quality so we are proposing to test for total coliforms, *E. coli* and the pathogens described below.

**Objective 2: Sample a subset of wells from the randomized sampling frame on a biweekly basis to assess seasonal variation in nitrate contamination.**

The flow characteristics of the Silurian dolomite aquifer, rapid transport from soil surface to the saturated zone and high groundwater velocities within the aquifer, can result in changes to groundwater quality that are rapid, intermittent, and short-lived.

This objective has two components: 1) installing a monitoring system on a non-pumping well that is capable of measuring water level, fluid temperature, and electrical conductivity and 2) sampling a sub-set of wells on a bi-weekly basis for nitrate-N.

We propose installing the monitoring equipment in a shallow monitoring well located in section 34 in the Town of Lincoln that was installed as part of Muldoon and Bradbury’s 2010 study. The monitoring data will be used to assess periods of recharge to the aquifer. Previous work (Bradbury and Muldoon, 1992, Muldoon and
Bradbury, 2010) suggests that water-quality parameters can vary significantly in relation to recharge. Having a continuous record of water levels, fluid temperature, and electrical conductivity will provide the necessary context for evaluating all of our sampling efforts.

We will randomly select, again stratified by depth to bedrock, 20 homeowners who will be instructed to collected bi-weekly samples. These bi-weekly samples will be collected by staff of the Kewaunee County Soil and Water Conservation Department and placed in a freezer. The samples will be retrieved on a quarterly basis and analyzed for nitrate-N. We anticipate that bi-weekly sampling would begin in October 2015 and continue through September 2016 such that there would be a full year of sample results.

**Objective 3: Sample the same subset of wells as described in Objective 2 for viruses and fecal markers capable of distinguishing septic versus bovine sources of contamination.**

We will sample the 20 wells described in objective 2 by ultrafiltration methods for human and bovine viruses and host-specific bacteria markers of fecal contamination. Nine hundred samples (450 wells X 2 events) will be collected during Objective 1 and 520 samples (20 wells X 26 sampling events) will be collected during Objective 2. For Objective 3 we propose sampling the same 20 wells once per season for a total of 80 samples.

Samples will be analyzed by qPCR (quantitative Polymerase Chain Reaction) for microbial targets divided into three fecal-source groups: 1) Human-specific microbes; 2) Bovine-specific microbes; and 3) Non-specific microbes found in fecal wastes of humans, bovines, and other animals. This latter group includes the human pathogens *Campylobacter jejuni*, *Salmonella* spp., enterohemorrhagic *E. coli*, *Cryptosporidium* spp. and *Giardia lamblia*. Samples will also be analyzed for total coliforms and *E. coli* by Quanti-Tray. Results from this sampling effort will be used to assess the sources of fecal contamination as well as the seasonal variability in pathogen occurrence in groundwater.

After completing study objectives 1, 2, and 3 groundwater quality data will be available for two large synoptic sampling events, temporal trends for a subset of wells included in the synoptic sampling, and pathogen and fecal source data for a subset of wells included in the synoptic and temporal sampling efforts.

**Objective 4: Install an automated sampling system on two wells in order to determine the timing of peak transport for viruses and indicator bacteria.**

Previous studies on groundwater sanitary quality in northeastern Wisconsin have relied on periodic monthly or quarterly grab samples (e.g. Masarik and Bonness, 2014; Borchardt et al. 2003). The flow characteristics of this aquifer --rapid transport from soil surface to the saturated zone and high groundwater velocities within the aquifer -- can result in changes to groundwater quality that are rapid, intermittent, and short-
lived. For this objective, we are proposing a sampling regimen better suited for capturing intermittent contamination events, leading to a more accurate assessment of the timing of pathogen contamination.

We will choose two domestic wells from the subset of wells chosen for bi-weekly sampling. Custom-designed automated samplers for pathogens and other microbes will be constructed by USGS personnel and installed to water taps in the basements of these homes. Based on previous research on recharge to the dolomite aquifer (Muldoon and Bradbury, 2010) we anticipate that there may be four to six discrete recharge events throughout the year. We will identify recharge events based on the monitoring parameters described in objective 2. We plan to sample these discrete recharge events as well as collect samples during periods without recharge. During recharge events, multiple separate samples will be collected at different stages of the hydrograph. During non-recharge events we will sample continuously over several days. Personnel from the Kewaunee Land and Water Conservation Department will retrieve samples within 24 hours of their collection and ship to Dr. Borchard's lab for analysis.

Samples will be analyzed by qPCR (quantitative Polymerase Chain Reaction) for the same microbial targets as described for Objective 3.

We anticipate that the auto-samplers will be constructed and installed in November of 2015 and that sampling will continue through October of the following year. The information gained from this objective is scientifically novel and will be crucial for developing effective sampling plans for future studies of the fractured dolomite aquifer.

**Objective 5: Perform statistical analysis of samples collected from this study and of existing water-quality data from Kewaunee County to assess spatial and temporal patterns of contamination.**

Existing water-quality data from Kewaunee County suggests that contamination of groundwater by nitrate-N and/or bacteria is not randomly distributed, but rather correlated with areas of thin soils (see figure 1). Analysis of existing water-quality data is always complicated by the fact that we do not know if the existing database is truly representative of groundwater conditions in an area. One could assume that the people submitting samples are those concerned about their water quality because of previously identified problems in the area or are well owners who have had bad samples results in the past (and thus the database is biased in terms of over-predicting instances of contamination). But it might also be that the majority of the samples are submitted by new homeowners, new parents who have new-found concerns about their water quality, or genuinely curious people (and the resulting database could be quite representative of current conditions).

This project includes a county-wide randomized and stratified sampling component. We can use the data collected from the random sample frame to explore the correlation between groundwater contamination (either by nitrate-N or bacteria) with both depth to rock and well casing depth. In addition, we can compare our randomized
sample results with the existing dataset at UW-Stevens Point Center for Watershed Science to assess whether the latter dataset is biased in terms of over-predicting instances of contamination. We will work in partnership with the Center for Watershed Science and Education to analyze the database of all existing water-quality sample results from Kewaunee County along with supporting data (location, known well construction reports, sample date, etc).

Our analysis of both the newly-collected sample results and the existing database of water-quality data will be based on existing maps of soil thickness in Kewaunee County and from the reported soil thickness on well construction reports (WCRs) for those wells that can be matched to a WCR. We are aware that the Wisconsin Geological and Natural History Survey (WGNHS) has begun a depth-to-bedrock mapping project for the Town of Lincoln, Kewaunee County, and possibly other regions of the county. We have excellent working relationships with WGNHS and if their new depth-to-bedrock maps are completed over the time-frame of this project, we will incorporate them into our analysis.

We are also aware that Mr. Joseph Baeten of the Wisconsin DNR is leading the effort for creating interactive GIS-based state-wide maps of agricultural fields covered under a nutrient management plan and approved for land-application of industrial, municipal, and septic system wastewater. The map for Kewaunee County is currently being updated and according to Mr. Baeton it will be available to us. If time and funding permit, we will explore the spatial associations between land use and private well contamination.

Seasonal variation in water quality will be assessed using the results from the subset of wells selected for bi-weekly sampling (Objective 2) and for virus/fecal marker sampling (Objective 4). Water-quality results from those subsets of wells will be compared to the data on water-levels and fluid temperature and conductivity data recorded at the non-pumping monitoring well (as described in objective 2). It is our hypothesis that nitrate-N values will tend to decrease during recharge events (due to dilution), whereas virus and bacteria counts will tend to rise during recharge events (as recharge will trigger transport from the soil surface).

**Deliverable Products:**

We anticipate that as for other Wisconsin DNR projects in which we have been involved, this project will involve a high level of communication, collaboration, and flexibility between DNR staff and the research team. Moreover, we are willing to serve on county or DNR organized committees and technical advisory groups related to the groundwater contamination issues in Kewaunee County. At present, we are anticipating the following project deliverables:

- Maps of results from the random sampling event(s) that will illustrate the spatial distribution of nitrate-N values and detections of bacterial contamination for Kewaunee County.
- Statistical analysis of the random sampling results that will assess correlation with soil thickness and well
• Statistical analysis of results from the 20 wells sampled biweekly for nitrate and quarterly for pathogens to assess seasonal variability in groundwater quality. The pathogen sampling results should also provide information on the sources of fecal contamination,
• Statistical analysis of results from the 2 wells sampled for viruses/fecal markers to assess the variability of pathogen transport in relation to episodic recharge events.
• Final report summarizing methods and results of the project.

Team Profiles
Dr. Randy Hunt is a Research Hydrologist with the U.S. Geological Survey in Middleton, Wisconsin. He completed his MSc and PhD at the University of Wisconsin-Madison. His areas of expertise focus on conducting research investigating groundwater flow and how it affects natural systems. This research program has used a variety of approaches such as numerical modeling, ion and isotope chemistry, parameter estimation, and stochastic methods. It has emphasized a range of groundwater–surface water settings including wetland, stream, and lake interactions. His work also includes investigation of ecohydrology, including the effects of groundwater on the biotic/ecologic community in aquatic systems.

Davina Bonness is currently the County Conservationist for the Kewaunee County Land & Water Conservation Department. She received her Masters of Science in Environmental Science and Policy from the University of Wisconsin-Green Bay and has been actively researching water quality, karst topography and land-use in Kewaunee County for the past 10 years. She is currently co-investigating the Intra-annual Variability of Well Water Quality in Lincoln Township to assist in analyzing groundwater quality and trends in Kewaunee County.

Laura Hubbard is a Hydrologist at the U.S. Geological Survey Wisconsin Water Science Center. As a member of the Toxics Substances Hydrology Program, she has been involved in multiple national studies investigating urban and agricultural contaminants in groundwater and surface water, including viruses and pathogens, hormones, disinfection by-products, and pharmaceuticals. She has expertise in site installation, instrumentation, and sample collection. Prior to the Wisconsin Water Science Center, Laura was a hydrologist at the Iowa Water Science Center where she studied nutrient and sediment retention in wetlands and virus and pharmaceutical occurrence in municipal groundwater.

Kevin Masarik works at the Center for Watershed Science and education as groundwater education specialist for the University of Wisconsin-Extension and University of Wisconsin-Stevens Point. He holds a B.S degree in
water resources from UW-Stevens Point and M.S. degree in Soil Science from UW-Madison. He assists Wisconsin communities in the promotion of well water testing and the collection and analysis of private well water data. He has worked extensively with well owners to assess groundwater conditions in Kewaunee County.

References Cited:


### Budget Details

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### Personnel Costs

| Portion of Dr. Muldoon’s summer salary ($5000) + fringe benefits (56.5%) | $7,825 |
| Minus USGS matching funds ($11,200) | $85,565 |
| We may be able to obtain the monitoring equipment from a USGS trial program ($4100) | $81,465 |
|  | **Total** | **96,765** |
|  | **Estimated total cost to DNR** | **$81,465** |

*Note: The USGS will provide a 20% match of the portion of the grant that goes to Dr. Borchardt’s lab (cost of virus sampling and autosamplers)
**Curriculum Vitae of Principal Investigators:**

Maureen A. Muldoon (Time spent on project, 25% of 12 months)
Professor, Department of Geology, University of Wisconsin Oshkosh
800 Algoma Blvd; Oshkosh, WI 54901-8649
Phone: (920) 424-4461; e-mail: muldoon@uwosh.edu

### Education
- Ph.D. Geology, University of Wisconsin-Madison (1999)
- M.S. Geology, University of Wisconsin-Madison (1987)
- A.B. Earth and Planetary Sciences (Cum laude), Washington University, St. Louis (1983)

### Experience
- 2014 - present  *Professor*, Department of Geology, University of Wisconsin Oshkosh
- 2004 - 2014  *Associate Professor*, Department of Geology, University of Wisconsin Oshkosh
- 1998 - 2004  *Assistant Professor*, Department of Geology, University of Wisconsin Oshkosh

### Research Interests:
Investigation of groundwater quality and flow in fractured dolomite, relationship between carbonate stratigraphy and hydraulic properties, and delineation of wellhead protection zones in fractured rock

### Professional Affiliations/registrations:
- Geological Society of America; American Geophysical Union; American Water Resources Association (Wisconsin Section); Association of Ground Water Scientists and Engineers; Licensed Professional Geologist and Professional Hydrologist in Wisconsin

### Relevant Publications:
- Bradbury, K.R., M.A. Muldoon, and J. Borski (2012) Monitoring Groundwater Inflow to the Mink River Estuary, Door County, WI. Final report to the Wisconsin Coastal Management Program (WCMP Grant Agreement Number: AD109367-011.03), 37 pages plus appendices
MARK A. BORCHARDT (Time spent on project: 20%)
USDA Agricultural Research Service, 2615 Yellowstone Dr. Marshfield, WI 54449
Telephone: 715-387-4943 Email: mark.borchardt@ars.usda.gov

Education

Professional Experience
Dr. Borchardt is a Research Microbiologist for the USDA – Agricultural Research Service and head of the Laboratory for Infectious Disease and the Environment, USGS, Wisconsin Water Science Center. He has 34 years of research experience, 21 years as a principal investigator, and he is an internationally recognized authority on the measurement, fate, transport, and health effects of human and zoonotic pathogenic microorganisms in the environment. He has written more than 80 scientific publications and been awarded over 50 research grants. Studies conducted by Dr. Borchardt have contributed to practices and regulations that protect the health and safety of millions of North Americans that use public drinking water supplies. He has given more than 150 invited presentations at regional, national, and international meetings, and has served on advisory panels for state and national scientific and regulatory organizations.

Refereed Publications Relevant to the Proposed Research