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**A STUDY OF FECAL INDICATORS AND
OTHER FACTORS IMPACTING WATER QUALITY IN
PRIVATE WELLS IN DOOR COUNTY, WISCONSIN**

by

Laurel A. Braatz

A thesis submitted in partial fulfillment of
the requirements for the Degree of

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in
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Approved:

Ronald D. Stieglitz
Major Professor

Ronald D. Stieglitz
Director of Graduate Studies

Committee Members:
John F. Katers
David M. Dolan

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Thank you to my niece for providing motivation by persistently questioning at every family gathering whether my thesis was complete. My niece was anxiously waiting for me to follow up on my promise to search for a puppy to adopt after I completed my thesis.

ABSTRACT

“A STUDY OF FECAL INDICATORS AND OTHER FACTORS IMPACTING WATER QUALITY IN PRIVATE WELLS IN DOOR COUNTY, WISCONSIN.”

Laurel Braatz

The primary goal of this study was to obtain the percentage of wells impacted by contaminants based on three indicators studied by the Environmental Protection Agency (EPA) as tools to identify fecal contamination in groundwater. The tests for *Escherichia coliform*, *enterococci*, and coliphage were conducted on 25 well sites in Door County, Wisconsin. This study provided insight on the impacts that the EPA Groundwater Rule (GWR) may impose on Door County public drinking water systems. The GWR will require systems that are vulnerable to fecal contamination in their source water to take actions to provide safe water. Actions to correct the problem include obtaining a safe source of water by utilizing an alternative safe water supply or by installing a water treatment system. Either solution to the problem could be costly. A secondary goal of the study was to identify a linkage between well water and illness in individuals consuming the water. The data that was gathered in this study is useful to regulators and lawmakers who are fine-tuning existing drinking water regulations and will implement the Groundwater Rule.

TABLE OF CONTENTS

ACKNOWLEDGMENTS	ii
ABSTRACT	iii
TABLE OF CONTENTS	iv
LIST OF TABLES	vi
LIST OF FIGURES	vii
CHAPTER 1 INTRODUCTION	1
Statement Of Problem	1
Purpose And Objectives Of The Study	2
Phase 1 – Monthly Testing Of 25 Wells	2
Phase II – Gastrointestinal Illness And Drinking Water	2
Study Site Rational	3
History Of Bacterial Contamination And Illness	4
Safe Drinking Water Act Groundwater Rule Background	12
CHAPTER 2 LITERATURE REVIEW	16
What Pathogens Are Found In Drinking Water	16
What Are Microbiologic Waterborne Illnesses?	17
Degree Of Illness Problems Caused By Contaminated Groundwater	17
Example Outbreak Cases	19
Illness Tracking Problems	21
Seasonal Contamination Of Drinking Water	24
Link To Precipitation	24
Factors Affecting Pathogen Transport	26
Links To Human And Animal Waste	30
CHAPTER 3 METHODS OF STUDY	32
Phase I Indicator Testing	32
<i>E.coli</i> And <i>Enterococci</i>	33
Coliphage	34
Gene Probe Detection Of Pathogens In Phase II Of Illness Related Water Samples	34
Data Analysis	36
CHAPTER 4 RESULTS AND DISCUSSION	38
Results Of 25 Wells Based On Rank	38
Analysis Of Indicator Tests	40
Results Of Indicator Tests Per Month For All Wells	41

Least Squares Regression Of Well <i>Coliform</i> Sampling With Environmental Factors	47
<i>Coliform</i> Count Relationship With Precipitation.....	58
Results Of Illness Testing.....	62
CHAPTER 5 CONCLUSTION AND RECOMMENDATIONS.....	65
REFERENCES CITED.....	70
APPENDIX A: WELL SITE AERIAL PHOTOS WITH KARST AND SOIL INFORMATION.....	75
APPENDIX B: WELL SITE GRAPHS OF <i>COLIFORM</i> AND PRECIPITATION VERSES DATE COLLECTED.....	101
APPENDIX C: WELL INDICATOR SAMPLING DATA.....	115
APPENDIX D: WELL SITE INFORMATION.....	124

LIST OF TABLES

4.1: Water Quality Rank Of 25 Well Sites.....	39
4.2: Pearson Correlation Matrix.....	40
4.3: Peninsular Research Station precipitation Data.....	42
4.4: Peninsular Research Station Temperature Data.....	44
4.5: Independent Variables Used In Model.....	50
4.6: Least Squares Regression Model With Year Round Precipitation Data.....	50
4.7: Regression Model Analysis Of Variance With Winter Data.....	51
4.8: Regression Parameter Estimates With Winter Data.....	51
4.9: Comparison Of Variables In Standardized Units.....	54
4.10: Least Squares Regression Model With Winter Data Removed.....	55
4.11: Regression Model Analysis Of Variance With Winter Data Removed.....	55
4.12: Regression Parameter Estimates With Winter Data Removed.....	56
4.13: Least Squares Regression Model With Winter Total Rainfall Term.....	57
4.14: Regression Model Analysis Of Variance With Winter Data Removed.....	57
4.15: Regression Parameter Estimates With Rainfall Data Summed.....	58
4.16: Precipitation And <i>Coliform</i> Graph Symbol Key.....	58

LIST OF FIGURES

1.1: Location Of Door County, Wisconsin.....	1
1.2: Wisconsin Groundwater Contamination Susceptibility Map.....	5
1.3: Well Casing Diagram.....	7
1.4: Door County Well Sites In Fecal Indicator Study.....	11
2.1: Door County Shallow Soils.....	27
2.2: Door County Karst Features.....	29
4.1: Percent Of Positive Indicators And Rainfall Per Month.....	43
4.2: Percent Of Positive Indicators And Rainfall Per Month.....	46
4.3: Door County Geologic Cross Section And Stratigraphy.....	48
4.4: Plot Of Regression Residuals Verses Depth.....	49
4.5: Plot Of Regression Residuals Verses Depth Without Well # 19.....	49
4.6: Door County Well Casing Requirements With Study Sites.....	53
4.7: Well # 15 <i>Coliform</i> And Precipitation Data Verses Date Sample Collected.....	59
4.8: Well Site # 15 Air Photo With Shallow Soil And Karst Information.....	61

CHAPTER 1 INTRODUCTION

Statement Of Problem

The proposed Environmental Protection Agency (EPA) Groundwater Rule will require public water systems that have been determined to contain fecal contamination to eliminate the problem with some corrective action (Blackburn et. al. 2004). In Door County, Wisconsin, the only solution for many water systems to obtain consistently safe water is to install costly treatment devices. Door County is located in Northeast Wisconsin (Figure 1).

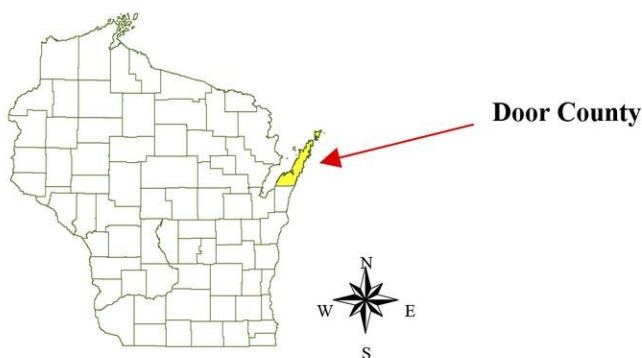


Figure 1.1: Location Of Door County, Wisconsin

The area is a popular tourist destination with an image as a pristine culturally blessed landscape. Unfortunately, the arts are not the only culture in abundance in Door County. *Coliform* are frequently found in water samples from public and private wells. Door County's fractured bedrock and limited soil cover make Door County more sensitive to intermittent groundwater contamination than many other areas of Wisconsin.

Purpose And Objectives Of The Study

Phase 1 – Monthly Testing Of 25 Wells

The primary purpose of this study was to sample 25 private wells monthly to obtain an average percentage of wells that would be impacted by fecal contamination based on positive detects in the three proposed indicator tests. The indicators include *Escherichia coliform* (*E.coli.*), *enterococci*, and coliphage. The EPA is considering use of these indicators to determine the water systems that would require treatment or a new water source to protect the health of the individuals these systems serve. Results for each test method were reviewed to identify similarities and differences in the detection of fecal contamination. Data collected from each individual study site were also studied to determine if any environmental factors were significant in explaining what wells were impacted.

Phase II – Gastrointestinal Illness And Drinking Water

The study also attempted to gather data on linkages between gastrointestinal illness complaints and the primary water source consumed by the individual. This data could then be used to teach others about drinking water problems and raise awareness of the health risks that exist.

Study Site Rational

The GWR will apply to all public water systems in Wisconsin. Door County is an appropriate location for this study for two reasons:

- (1) The area has a large number of small privately owned water systems.
- (2) The county is more susceptible to groundwater contamination than most other counties in the state due to the lack of soil and the vulnerable geology.

The size of a water system appears to be relevant to contamination occurrence. It is disturbing that data from the EPA show the highest occurrence of microbial unsafe water is found in small systems serving less than 500 people (Peterson 2001). Door County has only three community water systems with disinfection treatment equipment and trained operators to maintain the equipment and test the water quality. The majority of the remaining water systems in the county service less than 500 people and use untreated well water.

These small systems in Door County are also known to have a high occurrence of microbial unsafe water compared to other counties in the state. Analysis of Transient Non-community (TN) wells within the twelve county area in the Northeast DNR region showed that Door County had significantly more confirmed *coliform* unsafe wells than the other counties based on an equal probability of well contamination (Hodgson 2002). A TN is a system that serves at least 25 people at least 60 days a year but there are less than 25 of the same people served more than 6 months. TN systems include churches, campgrounds, restaurants, and motels with individual wells.

Private home wells were utilized as sampling sites in this study. Wisconsin well construction standards for private homes and TN wells are both regulated by Wisconsin Administrative Code NR 812 (WI DNR⁵ 2001). The sampling data gathered in this study from private wells should be representative of TN public wells since the same standards for well location, and well construction methods apply to both types of wells.

History Of Bacterial Contamination And Illness

Door County exhibits a long history of contaminated water wells. The area is more susceptible to groundwater contamination than most other areas in the state. A study of Wisconsin identified many areas in the county as some of the most susceptible areas for contamination in the state (Figure 1.2).

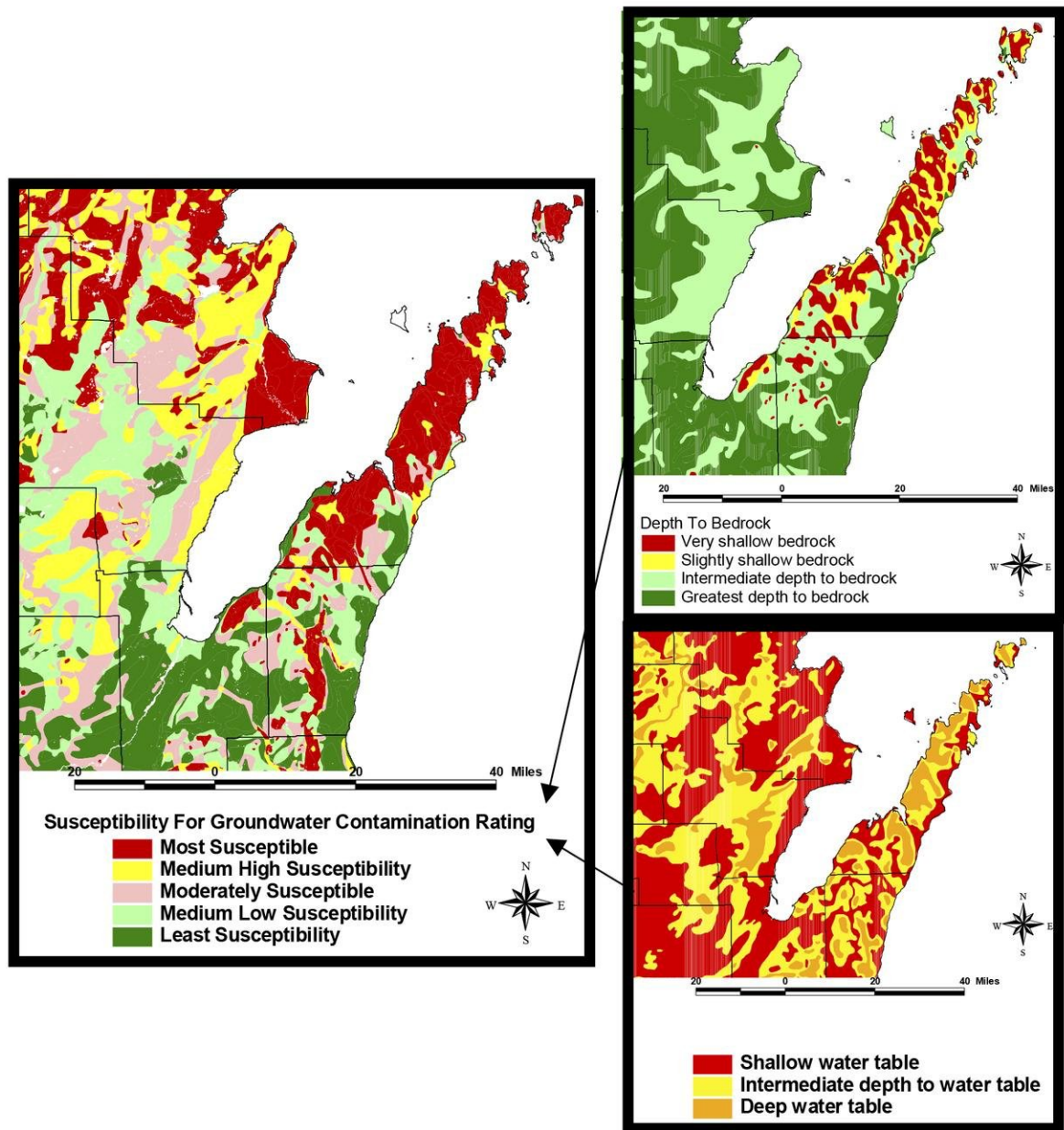


Figure 1.2: Wisconsin Groundwater Contamination Susceptibility Map¹ (WI DNR⁴ 1989).

¹The model used to create the susceptibility map utilized depth and type of bedrock, depth to water table, soil depth and type, and surficial deposit data to identify general areas of concern. The data should not be used for site specific analysis.

Door County is the only county in the state of Wisconsin that currently requires quarterly *coliform* bacteria sampling for TN water systems. In 1993 heavy precipitation events contributed to numerous *coliform* positive tests for wells in the Village of Ephraim. Testing of 33 public and private wells during 1993 found 17 of the wells were contaminated with *coliform* bacteria. In response to the unsafe *coliform* positive water samples and cryptosporidium found in an Ephraim TN water system in 1994, the entire county was placed on quarterly sampling (WI DNR¹ Unpublished Door County Files). The Village of Ephraim may have spotlighted the groundwater contamination problem, but much of Door County faces similar problems. Most of Door County contains little soil on top of fractured dolomite bedrock. Therefore, most of the county is highly susceptible to groundwater contamination.

Groundwater contamination in Door County has been documented as a problem since 1916. Typhoid outbreaks occurred in Sturgeon Bay in 1918, 1921, and 1922. The source of the outbreaks was traced to contaminated private wells (Wisniowski 1942). In 1924, the Sturgeon Bay public water supply was contaminated due to leakage from the sewer system. In 1927, another typhoid outbreak occurred in the Baileys Harbor area. Heavy rains and pit toilets located on top of fractured bedrock were believed to have caused the contamination. In 1929, an orchard located in the Town of Sevastopol experienced an outbreak of typhoid. The contamination event was blamed on a poorly constructed well. Gastrointestinal illness has also been prevalent in Door County's history. Cases were documented at migrant camps in 1939 and 1940 (Wisniowski 1942).

Water quality studies conducted in Door County by the Health Service in 1955 and 1957 continued to identify contaminated water supplies. In response to these data the well construction requirements for the county were made more stringent. Well casing depth amounts were increased from the minimum amount of 40 feet to a minimum amount of 100 feet to provide greater protection from surface contamination (WI DNR² 1957). Well casing is the pipe installed in a well bore hole and grouted in place to prevent shallow groundwater from above the bottom of the pipe from gaining access to the well (Figure 1.3).

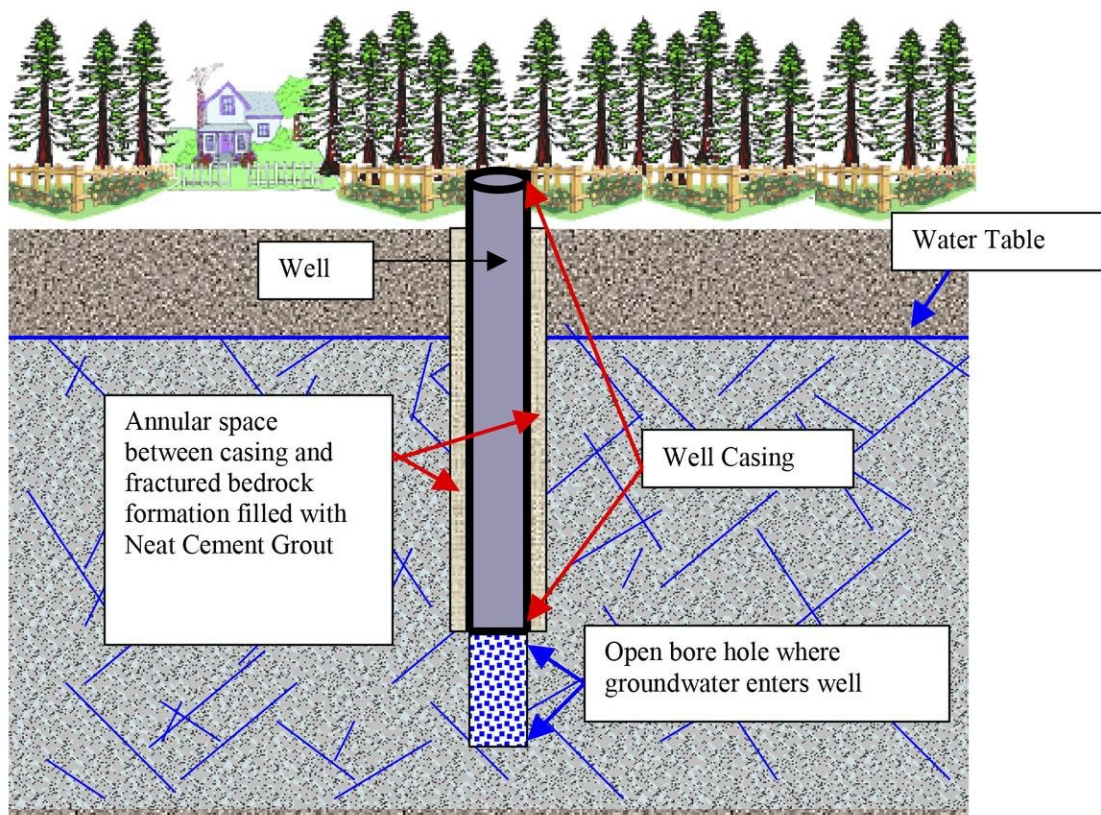


Figure 1.3: Well Casing Diagram

Contamination of the water supplies in Door County remained a problem even after the new casing depth requirements were implemented. The contamination continued for at least two reasons: (1) Older shallow cased wells remained in use and (2) newer wells were being contaminated in some areas despite the additional protection provided by the increased casing depth.

In 1968 there were 44 cases of hepatitis and a death was linked to consumption of contaminated drinking water on Washington Island. Heavy spring rains had washed septic system waste into the groundwater system (Wausau Daily Herald 1968). In response to this illness outbreak incident, a study was undertaken by some Door County residents in 1970 to draw awareness to the continuing problem. The study showed continued contamination of Door County wells and coined the phrase “Poison in Paradise.” The study did suggest wells with 100 feet of casing provided better quality water (Olesen 1971).

The state geological survey conducted a study of the Door County geology and wells in 1971 in response to the continued concern over the drinking water quality. The data gathered was used to establish new casing advisory areas based on the groundwater, soil and bedrock information. The new minimum casing depth in sensitive areas was increased to 170 feet (WI DNR³ 1971). The casing requirements established in the fall of 1971 for Door County remain in effect today.

One again, stricter casing requirements did not eliminate contamination problems in the county. From 1987 to 1996 Door County was the only county to have more waterborne illness outbreaks reported than foodborne outbreaks (WI DHSS 1998).

Documented outbreaks of waterborne illness occurred in Ephraim in the 1980s and 1990s. In 1988 a restaurant water supply in Ephraim was contaminated with sewage. There were 37 people confirmed to be ill out of the 64 people (58%) that were contacted. Based on the information gathered, the Department of Health estimated 340 people out of the 1000 people exposed became ill. In 1989 a motel water supply well was contaminated with fecal material from the pressurized sewer line. There were 6 people confirmed to be ill with gastrointestinal illness symptoms. In the spring of 1997 a restaurant in Fish Creek experienced an illness outbreak. The following week food and water samples collected by the Department of Health tested free of bacterial contamination. Statistics on the affected individuals health surveys suggested a link to the pasta, which would have been rinsed in water from the well and later placed in a food warmer (WI DNR¹ Unpublished Door County Files). The randomness of contaminants passing through the aquifer could explain why a water sample collected the following week failed to identify indicator bacteria.

Failure of grinder pumps in the village of Ephraim continued to put wells at risk in the 1990s through 2003. A portion of the Ephraim collector sewer is pressurized to force sewage to the upgradient sewage treatment plant. A loss of pressure was created by the failure of a grinder pump check valve on private property. This provided a pathway for

the village sewage downgradient of the grinder pump to escape from the collector sewer through the private piping and flood the property. The spilled sewage seeped into the ground and contaminated onsite wells. Wells with fecal contamination were identified in 1997 and 2003 as a result of check valve failures (WI DNR¹ Unpublished Door County Files). Fortunately, no illness outbreaks were documented from these occurrences. However, the events show that the vulnerability of water supplies in this county to contamination from surface fecal material continues. Steps such as new testing methods or installation of disinfection equipment need to be taken to make water supplies safer.

The EPA's strategy in the proposed GWR and existing Safe Drinking Water Act requirements is to prevent groundwater contamination and illness outbreaks from occurring. Testing to identify wells vulnerable to fecal matter at the source is one method the EPA hopes will aid in finding problem water systems before an outbreak occurs. This study of 25 Door County sites provides a pre-view of the percentage of "sensitive" wells that may be identified as contaminated by the proposed GWR testing.

The 25 well sites were selected from a group of private well owners that volunteered to be in the study. The volunteers were narrowed down to the 25 sites based on location and well construction information (Figure 1.4).

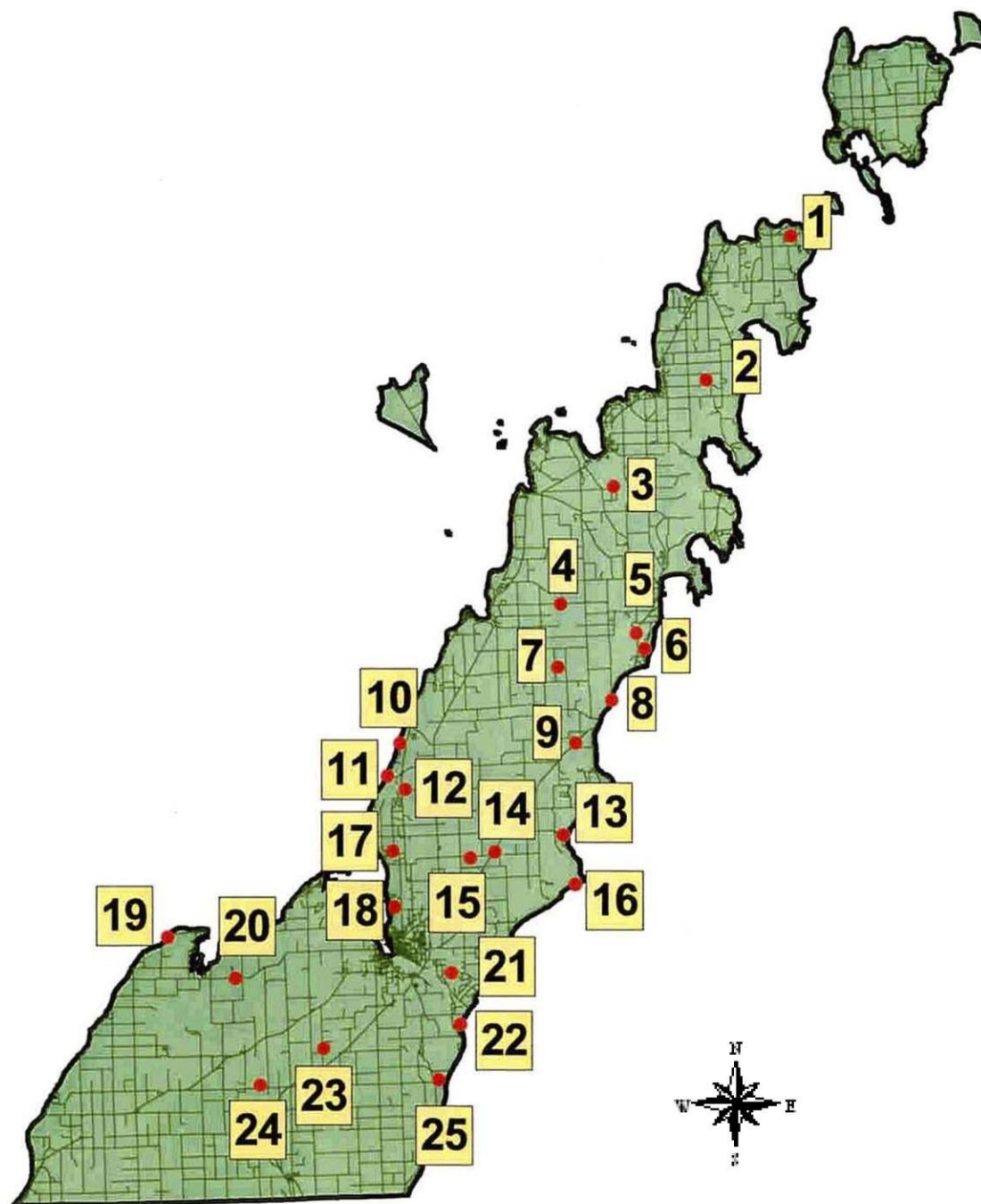


Figure 1.4: Door County Well Sites In Fecal Indicator Study

Safe Drinking Water Act Groundwater Rule Background

The Safe Drinking Water Act (SDWA) enacted in 1974 provided authority to the EPA to regulate drinking water. The SDWA made the supplier responsible for the quality of the water they served. In 1996 the EPA was directed to develop rules to require disinfection of groundwater, when needed, to protect public health (USEPA¹ 2000). The GWR was proposed in 2000 and developed in response to this amendment. The EPA is expected to release the final version of the rule in 2005. The GWR will include five preventative strategies that prior EPA drinking water legislation did not adequately address. The first aspect includes sanitary surveys of public systems to identify deficiencies. The second aspect is a hydrogeologic sensitivity assessment of each public system to identify wells sensitive to fecal contamination. The third aspect is source water monitoring. Currently, the Safe Drinking Water Act focuses on sampling for microbial indicators in the distribution system. The fourth aspect of the rule will require corrective action for non-complying features found in the water system and eliminating fecal contamination with treatment or providing an alternative permanent source of water. The fifth aspect of the rule is monitoring requirements to ensure that treatment equipment is maintained (USEPA¹ 2000). Wisconsin already conducts inspections and requires correction of non-complying features. Therefore, the major impact to Wisconsin of the proposed EPA GWR will be additional monitoring of source water for sensitive systems and the wells found to contain fecal contamination will require installation of approved treatment devices or the use of a new water source.

Due to the presence of karstic bedrock and shallow soils, all Door County public water systems will likely be classified as being located in a sensitive geologic area under the second aspect of the GWR. Classification as a sensitive system will lead to an increase in required monitoring. The additional monitoring proposed in 2000 would require sensitive groundwater systems to collect a minimum of 12 consecutive monthly samples for fecal indicators. The EPA GWR promulgation team has been studying the use of culture tests for *enterococci* bacteria and male specific coliphage viruses, in addition to the *Escherichia coliform* (*E.coli*) bacteria test (already required under existing SDWA rules) for early detection of groundwater fecal contamination (Smith 1997). The EPA will decide which fecal indicators will be required based on studies of different methods.

The EPA has proposed that wells identified as fecally contaminated, under the GWR, may be required to install costly water treatment devices or obtain an alternative safe source of water. Door County does not have a feasible alternative source for obtaining drinking water in most areas of the county. Therefore, the only solution for many Door County systems is to install an approved water treatment device. Approved devices for microbiological disinfection include (1) ultraviolet lights with pre treatment filters and softening and (2) disinfection with chlorine followed by carbon filtration to remove excess chlorine. Both approved device installations require monitoring equipment to constantly determine whether the treatment equipment is properly functioning. The upkeep of the equipment and/or replacement costs for lights and filters are a significant investment that continues beyond the initial price to install the treatment device.

The cost to install treatment and monitor treated water quality may have undesirable economic consequences on small businesses, community buildings, and churches that own water systems. EPA data suggest implementing the GWR would cost the average system serving less than 100 people \$191.87. The estimated cost per household was estimated at \$19.37 (USEPA¹ 2000). However, these costs are averages for the entire country. The estimates include wells in aquifers that are more stable and may not require any treatment.

A TN system in Door County that recently obtained a bid for ultraviolet treatment on one well that served approximately fifteen single family condominium units was quoted a price of \$46,000. This did not include annual maintenance or light and filter replacement (WI DNR¹ Unpublished Door County Files). Door County well owners are interested in providing safe water. However, well owners want information to justify the need to install treatment units and are concerned about the high cost that a small water supplier would have to pay.

One deficiency of the current State Revolving Loan Fund for installing treatment equipment or upgrading water systems is that small systems like the TNs in Door County are not eligible for the money. Systems allowed to obtain funding must meet the definition of a community system (USEPA² 2000). A public water community system is defined as serving the same 25 people all year. The TN public systems do not meet this definition because they may be seasonal or do not serve the same people year round. It is a major challenge for a small TN system such as a campground to find \$10,000 to

\$50,000 for the cost of a treatment system. Political lobbying by smaller impacted parties may be the only mechanism to motivate changes to the laws that govern what types of systems are eligible for loans.

The financing for installing water treatment systems for wells with known groundwater contamination is not the only challenge to provide safe drinking water. In addition, the present microbiological monitoring program does not adequately identify contamination episodes in the water system. The Safe Drinking Water Act (SDWA) only requires TN systems to monitor once per year for *coliform* with a waiver or quarterly without a waiver¹, and collect a nitrate once per year (WI DNR⁶ 2003). Groundwater wells have been monitored for *coliform* and *E. coli* bacteria to determine potability of water supplies for more than 100 years (Grimes, 1999). The *coliform* group does contain *E. coli* bacteria, which is indicative of fecal contamination. However, *coliform* testing may also detect nonpathogenic microorganisms that may have originated from sources other than animal or human waste. Therefore, water quality monitoring that relies exclusively on traditional *coliform* testing from the distribution system may result in positive results that do not pose a health risk. Another failure of the standard indicators required by the current Safe Drinking Water Act is that they cannot accurately predict protozoa, viral, or non-*coliform* group bacteria pathogens. The GWR is an additional attempt to better diagnose water systems that do pose a health risk to consumers and prevent unsafe water from being served to consumers.

¹Door County is the only county in Wisconsin that is required to monitor quarterly and not eligible for a waiver because of past sampling data that showed the area was vulnerable to groundwater contamination.

CHAPTER 2 LITERATURE REVIEW

What Pathogens Are Found In Drinking Water?

Bacteria, viruses, and protozoa have all been found in drinking water and have contributed to illness outbreaks. Viruses are the smallest pathogen and are found in a size range of 1 – 100 nanometers (nm). Bacteria by comparison are 30 to 10,000 nm. Protozoan are the largest pathogens of concern in drinking water at 1000 to 10,000 nm (Clear Tap 2002). Virus pathogens are the smallest, most resilient, and require a low ingested dose for infection. EPA estimates that for every five cases of gastrointestinal illness caused by viruses there is only one caused by bacteria (Peterson 2001). A recent study of 50 wells in Wisconsin that were sampled quarterly found viruses in four wells. The two Door County wells in the study both contained viruses in at least one quarterly time period. The only well in the study to have more than one virus identified throughout all quarters of sampling was also one of the Door County wells (Borchardt¹ et. al. 2003). Despite these data that suggest other non-*coliform* pathogens are found in drinking water and may cause illness outbreaks, the only microbiological monitoring in place for groundwater is *coliform/E.coli* sampling. This suggests that while huge strides have been made from the days of frequent waterborne outbreaks in the United States there are still more improvements in testing needed to ensure safe drinking water.

The EPA and American Water Works Association Research Foundation are conducting studies to determine the occurrence of the proposed fecal indicators using different testing methods (Smith 1997). The focus on better fecal indicator testing of groundwater

at the source that supplies drinking water may help prevent additional illness outbreaks due to pathogens, without falsely alarming the public.

What Are Microbiologic Waterborne Illnesses

There are a variety of illnesses that sensitive individuals can become infected with. The most common symptom of consuming water contaminated with fecal material is gastrointestinal illness. Symptoms may include the following symptoms: headache, nausea, diarrhea, upset stomach, and cramps. However, pathogens can cause other illnesses. Some of these diseases are chronic rather than acute and therefore difficult to associate with contaminated drinking water. Coxsackie B virus is believed to cause insulin dependent diabetes. *Campylobacter* bacteria is believed to cause paralysis through Guillian-Barre syndrome (Peterson 2001). *Campylobacter* bacteria is also associated with long term joint pain and illnesses in the bladder, urinary tract, or liver. It is estimated that 2-10% of people who contract *E.coli* 0157 may experience long term health problems caused by the organism. Drinking *E.coli* 0157 contaminated water may cause chronic health effects like irritable bowel syndrome (Simone 2000). Some viral pathogens are linked to myocarditis – an inflammation and degeneration of the heart muscle (USEPA² 2000).

Degree Of Illness Problems Caused By Contaminated Groundwater

There is an increasing concern among scientists and water providers over microbiological contamination. Data on waterborne outbreaks suggest that the number of human deaths from microbes in water is higher than the number of cancer deaths caused by chemicals

in the water (Berger 1995). The Centers for Disease Control and Prevention (CDC) estimate 900 to 1,000 people die from microbial illnesses associated with groundwater contamination each year in the United States. Contaminated water is estimated to cause gastrointestinal illness in 7 to 30 million Americans each year (Gelt 1998). In approximately half of the documented community illness outbreaks in the United States over the past 25 years, an etiologic agent has not been identified (Craun 1988;1992). These data suggest current water supply monitoring practices do not appear to be adequately protecting water consumers.

Hospital data support the need for better management and protection of drinking water. A study of elderly people in Philadelphia found that at least 10% of hospital admissions were related to drinking water even though the treatment plant met current standards. There was an association with increased illnesses within a week of an increase in the turbidity (Herman 1999). A similar study in children found that 10% of children that required hospitalization were linked to illnesses caused by drinking water (Mercola 1997).

The actual numbers of reported and documented illness cases in a community are far below the percentages listed above. The illness outbreak cases that are documented usually involved a large population of people exposed to the contaminated water. Many smaller waterborne illness outbreaks don't attract the press or may not even be reported. Even with the low rate of reporting the documented illness cases raise concerns that better protection of drinking water is needed.

Example Outbreak Cases

The Washington County Fair, New York, made headlines in September 1999 when a drinking water well became contaminated with *E.coli* 0157 and *Campylobacter jejuni*.

At least 781 people became ill, 127 cases of *E.coli* 0157 were confirmed and 45 cases of campylobacteriosis were identified. There were two people that died as a result of consuming contaminated drinking water (New York State Department of Health 2000).

The following year in May of 2000, the entire population in the Canadian town of Walkerton was exposed to *E.coli* 0157. The town's water system had been testing unsafe for weeks but there was no action taken by utility employees. Negligence of the water service providers resulted in the death of seven people (Ahluwalia 2002).

An outbreak of *Campylobacter jejuni* that occurred in Green Lake, Wisconsin, in January of 2000 at an Amish farmhouse is one example of a smaller illness case. A group of 18 people visiting the area stopped for a scheduled Amish lunch. There were a total of 13 people that became ill. The water supply was tested after the outbreak and reported to be unsafe. An investigation of the site suggested a chicken coop located beside the well was the source of the contamination to the well (Archer 2001).

Bacteria are not the only pathogens that have been documented as waterborne illness outbreaks. Waterborne viruses which current water testing methods do not detect have also been confirmed to sicken individuals. Norwalk like virus was identified as the cause of groundwater illness in a 1997 outbreak at a New York ski resort. A total of 1,450 people became ill from consuming ice made from the water supply. In July of 2000, a

Norwalk like virus illness outbreak occurred at a campground in California. A total of 147 people became ill during this event. Another Norwalk like virus outbreak occurred in Kansas in June of 2000. A total of 86 people that had attended different events at a reception hall became ill (Lee et. al. 2002).

The limits of standard *coliform* testing to predict water contaminated by protozoa are also a problem. Protozoa are another waterborne threat to human health. These pathogens are generally associated with surface water systems like the Milwaukee, Wisconsin, outbreak where 403,000 people became ill from consuming cryptosporidium in the municipal drinking water supply (USEPA¹ 2000). Groundwater is normally thought to provide adequate filtration that is capable of removing the protozoa due to their relative large size for a groundwater pathogen. However, cases documented by the Center for Disease Control (CDC) suggest that protozoa also pose a threat to drinking water from groundwater sources. In 1999 and 2000 there were six occurrences of groundwater sources for drinking water being contaminated by either girardia or cryptosporidium (Lee et. al. 2002). Door County has had at least two wells confirmed to have contained cryptosporidium based on laboratory testing (WI DNR¹ Unpublished Door County Files).

An outbreak on South Bass Island, Ohio, in August 2004 involved all three types of pathogens (bacterial, viral, and protozoan). The CDC assisted the local EPA with the outbreak investigation. Initial data confirmed fifteen illness cases caused by campylobacter, one illness caused by salmonella, seven illnesses caused by norovirus and one illness due to giardia. Based on personal interviews with ill individuals, a total of

1,377 people were believed to have become ill from consuming the contaminated drinking water on the Island (Smith 2004).

This South Bass outbreak provides a warning of what could happen in Door County. The two locations do have similarities. The drinking water on South Bass Island is obtained from the same fractured dolomite formation that makes up Door County's major aquifer. Similarly, South Bass Island has thin soil cover (only six inches in some areas) (Murphy 2004). Both areas also rely heavily on tourism to support the local economy and illness outbreaks can have disastrous impacts on the tourist industry.

Illness Tracking Problems

The outbreak examples presented show there is a continuing problem with waterborne illness. People are becoming ill from drinking water and experiencing both health and financial impacts. Why aren't more people complaining and requesting that more be done for prevention? One answer to this question is the difficulty in diagnosing and documenting illness outbreaks. The mobility of people consuming the contaminated drinking water and lack of knowledge of illness symptoms complicate waterborne illness outbreak identification. The transient nature of people consuming water at motels, restaurants, and campgrounds that predominate the Door County landscape, means people may travel out of the area before illness symptoms are identified. Illness victims from the same water source may have returned to their homes and be miles apart. A linkage to the individuals common illnesses source may not be made as a result of the traveling.

Confusion may also occur because only a small percentage of people are impacted by most outbreaks. Some people have a natural tolerance to the contaminants or may have acquired immunity to a pathogen from prior exposure. Therefore, those people who do become ill may not link the illness to the water source because other individuals who consumed the water did not become ill. Researchers estimate that up to 20-25% of the US population is sensitive to illnesses from drinking water due to their age, and/or medical condition (Gerba et al. 1996). The variance in people's immune systems can yield different results in people becoming ill from the same dosage of a pathogen. The health standards that EPA sets for contaminants are established to protect this vulnerable subset of the population. Therefore, water identified as "unsafe" may not cause illness in the majority of the people exposed even if it does contain pathogens. For example, a family of four, camping, may only have one individual become ill and the connection of the illness to drinking water may not be made.

Another problem in identifying waterborne illness is that most cases are not reported. Data suggest that only 5% of bloody diarrhea cases are reported and less than 3% of the non-bloody diarrhea cases are reported (Kramer et. al. 2001). Usually, people's immune systems fight off the problem in a few days. People do not prefer to discuss their bodily functions unless the problem persists longer than a few days. In this study it was difficult to get volunteers to donate specimens during the time that they were experiencing diarrhea.

The intermittent and inconsistent distribution of contamination in water systems further complicates identifying waterborne outbreaks. People do not always receive an effective dose of the pathogen so all people who consume the same water may not show symptoms. Door County TN systems often do not exhibit the same results in all four 100 mL sample bottles collected for follow up to an unsafe sample, even though the samples are collected immediately after each other. The mixed results show the bacteria are not evenly mixed in the water supply. Therefore, even people consuming water from the same source may not be exposed to the same concentration of pathogens.

The symptoms of waterborne illnesses are also difficult to distinguish from other ailments. The most common symptoms include nausea, headache, upset stomach, cramps and diarrhea. There are many other potential causes for these symptoms and if only a few people become ill from drinking contaminated water it is easy to blame other sources for the illness. The symptoms experienced by each person consuming contaminated water can also vary. One person's headache may not be linked with another person's diarrhea.

For the reasons discussed in the prior paragraphs many contamination events with small numbers of people exposed are often missed. Milwaukee, Wisconsin, had one of the largest waterborne illness outbreaks with 400,000 people that became ill from cryptosporidium. It was only after half of the people became ill that the drinking water link was looked into (Kramer et. al. 2001). A WI DNR employee made the comment that it took a pharmacist reporting the disappearance of "Pepto Bismol" off the shelves before

the waterborne illness problem was identified. This doesn't provide much faith that a smaller event would even be noticed. Additional study of the connections between drinking water and illness is needed to better identify outbreaks and to prevent them from occurring.

Seasonal Contamination Of Drinking Water

The time of the year is believed to influence when contamination events occur. Based on previous research by the Wisconsin State Laboratory of Hygiene (SLOH) and observed increases in TN *coliform* positive monitoring results, there are more bacterial unsafe wells in the months of July – October. The bacterial seasonal occurrence may be due to temperature changes. The Wisconsin SLOH conducted a study of 65 water systems and found a positive relationship between the temperature of the water and *coliform* positive results (Olstadt et. al. 1999). Studies of enteroviruses and hepatitis E also show a seasonal trend as these viruses are more prevalent in fall and winter (Warrington 2001). Data from the CDC for all waterborne illness outbreaks show the highest occurrence of cases are reported during the months of June through September (Blackburn et. al. 2004). This makes sense if the greatest number of bacteriologically contaminated wells occurs in combination with some of the viral contaminants during this period.

Link To Precipitation

Another factor believed to cause contamination of drinking water is precipitation events. Analysis of illness outbreak data by Johns Hopkins University linked waterborne disease outbreaks to recent heavy rainfall events in more than 50% of the documented cases. An

outbreak is defined by at least two individuals with the same illness. Groundwater contamination correlated with a significant rainfall event within the past three months (Parsons 2001). Data suggest Door County contamination would likely have a quicker response than three months based on the lack of soil and fractured dolomite bedrock. Work conducted by Ken Bradbury and Maureen Muldoon for the Wisconsin Geological and Natural History Survey showed changes in well water temperature and electrical conductivity within 24 hours of a rainfall event. The temperature and electrical sensor was located in a Sturgeon Bay well at a depth of 250 feet. The well was cased and grouted to 170 feet (Muldoon 2001). The minimum groundwater flow rates in the vicinity of Sturgeon Bay, Wisconsin, were estimated at 13-115 feet per day (Bradbury et. al. 1997). Groundwater in more stable aquifers may only move inches per day. In comparison, the Central Wisconsin Sands aquifer may flow 1 to 5 feet per day (Schmidt 2004). However, in Door County flow rates ranging from 55 feet to 280-300 feet per day have been suggested in some groundwater studies (Muldoon et. al. 1992).

The faster impact of rainfall on Door County wells is also supported by the general observation that more TN wells test positive for *coliform* bacteria after recent rainfall events. A frequent comment heard from transient non-community samplers in Door County is “the unsafe water sample is due to the recent rain – I don’t believe there is anything wrong with the water” (WI DNR¹ Unpublished Door County Files).

Surface water often makes the news when contaminated with sewage, fertilizer, or animal waste but often groundwater is thought to be safe because it mysteriously comes from

some unseen place below ground. The linkage between contamination in surface water and the groundwater quality below are often overlooked.

Factors Affecting Pathogen Transport

While groundwater is naturally filtered through native soil and rock formations, not all areas were created equal with regards to filtering ability. The amount of soil and type of soil located in an area affect the natural filtering ability of a section of land. There is a greater chance that surface contamination will be flushed into the aquifer below a site if little soil exists. Shallow soil areas are present in much of Door County and are believed to have an impact on water quality in local aquifers (Figure 2.1).

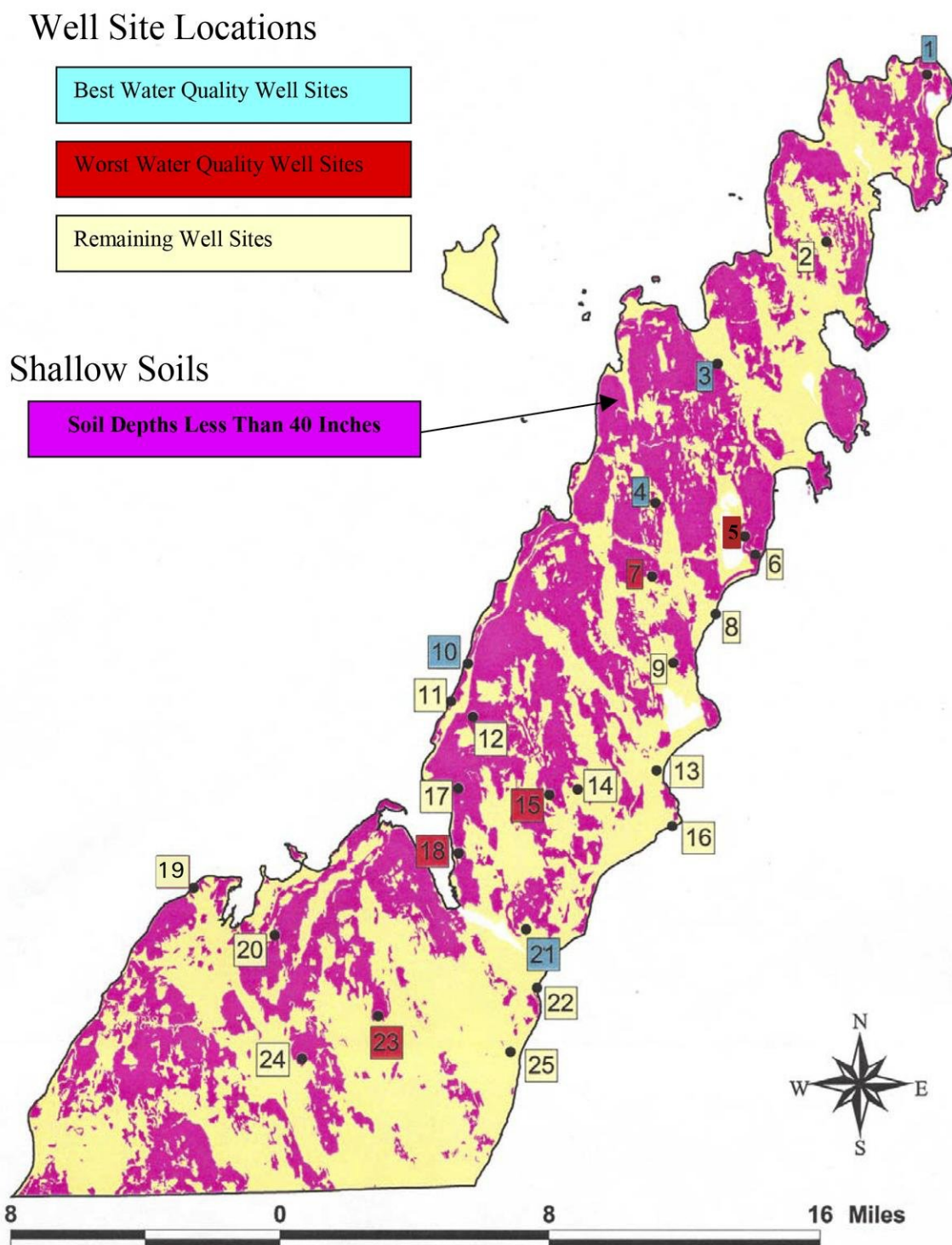


Figure 2.1: Door County Shallow Soils (USGS 1992)

Karst, fractured rock and gravel areas are identified in the GWR as hydrogeologic settings that are sensitive to fecal contamination. Only 15% of all public systems are expected to fall into this category (USEPA¹ 2000). However, most of the Door County water systems will be defined as sensitive. Sensitive areas lack a hydrogeologic barrier to prevent surface contamination from entering the aquifer below. Sensitive areas also allow groundwater to flow rapidly due to large voids in the aquifer. Door County's main aquifer consists of fractured dolomite. Door County has many shallow karst features that allow rapid movement of water into the aquifer below. Karst features are created by dissolution of the limestone/dolomite rock formations by a weak acid created from a chemical reaction between rain and carbon dioxide. Karst features include fractures, caves, sink holes (dolines) and swallets. DNR geographic information system data layers exist for karst features in Northern Door County and portions of Southern Door County¹. The map created for Door County with this data provides an idea of areas where karst features are prevalent (Figure 2.2). Areas with greater numbers of shallow karst features will allow surface water carrying fecal material to enter the groundwater aquifer and may impact drinking water wells withdrawing water from that aquifer.

¹The karst features in Southern Door County were not inventoried as extensively as Northern Door County. Southern Door County fractures were traced from air photos in the Red River Watershed and nearby areas. Many more karst features may exist throughout the county but have not been mapped.

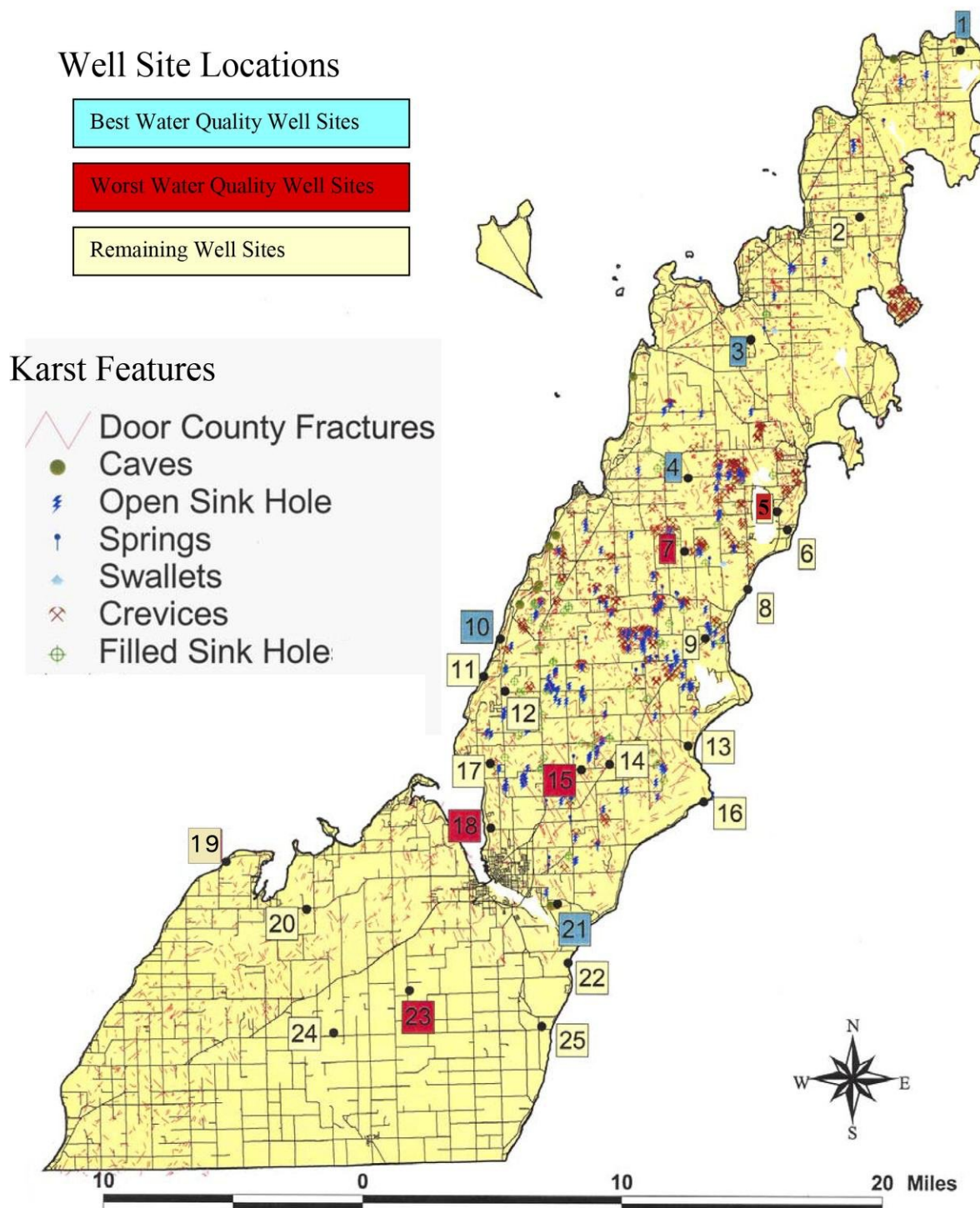


Figure 2.2: Door County Karst Features (Stieglitz 1986, Stieglitz 1994)

Links To Human And Animal Waste

Any surface contaminant that can be carried into the aquifer through a precipitation event is a threat to drinking water. The high flow rates for Door County aquifers allow untreated or partially treated septic wastes to infiltrate the groundwater quickly. Even functioning septic systems may have piping leaks or the final effluent may still contain pathogens. It is estimated that 40% of groundwater illness outbreaks are related to on site septic failures (Meschke et. al. 1999). The EPA estimates that groundwater contamination may occur if there are more than 30 septic systems per square mile. A study in central Wisconsin focused on holding tanks and found an 8% increase in viral illness with each additional holding tank per square mile. There was a 22% increase in bacterial illness for each additional holding tank per town quarter, quarter section (Borchardt² et. al. 2003). This study did not evaluate other types of septic systems but it is believed they would likely be more of a concern because the waste is treated on site. In addition, the older failing septic treatment units are bigger concerns for release of pathogens into the aquifer. The Door County Sanitarian office data show an average failure rate of 25% in septic systems inspected at the time of sale (Teichtler 2004). Leaks and effluent from functioning septic systems along with the systems diagnosed as failing, all contribute to groundwater contamination. The private septic waste is not the only source of human waste of concern in Door County.

Newer, poorly designed sewer systems and aging community sewer systems are also believed to have contributed to groundwater contamination in Door County. The Ephraim sewer system has had numerous spills on private property due to the failure of

check valves on private grinder stations. One spill dumped several thousand gallons of sewage on properties with bedrock at a few feet (Kincaid 2003). Two illness outbreaks in the late 1980s occurred at sites where spills from the Ephraim sewer system occurred (WI DNR¹ Unpublished Door County Files). Wells in the Village of Valmy were sampled in 1998 and 10 out of 11 had been contaminated with *coliform* and three wells contained fecal material (WI DNR¹ Unpublished Door County Files). The village has an older gravity sewer system. The source of the well contamination was either from leaks in the sewer system or from a nearby farm and farm fields. The WI DNR Wastewater Engineer inspected the sewer and the WI DNR Animal Waste Specialist inspected the local farming operation but neither could be directly linked to the problem. There is also evidence of wells being impacted near manure spreading locations. Two tavern wells in Door County, were confirmed to contain fecal material, shortly after adjacent farm fields were spread with manure (WI DNR¹ Unpublished Door County Files). Some wells included in this study were located in active farming areas and were suspect to have been impacted by contamination from farming sources.

CHAPTER 3 METHODS OF STUDY

Phase I Indicator Testing

An advertisement in a Door County paper and pamphlets dispersed at community meetings were used to solicit study participants. Volunteers submitted information sheets on site location and well construction. Each site selected had to be able to provide year round access to the water system for sampling. Well sites were selected from the group of volunteers to disperse sampling sites across as much of the county as possible.

Participants were also screened based on the amount of well construction information available. All of the wells selected to be in the study did have well depth information available. Well sites were also selected to include a diverse grouping of casing depths.

The 25 wells were sampled monthly for a year to capture seasonal variability. This sampling frequency also matched the proposed schedule in the GWR for sensitive systems. WI DNR staff collected water samples from the well sites that were included in the study. The Wisconsin State Laboratory Of Hygiene (SLOH) analyzed the water samples for *coliform* bacteria and fecal indicators (*E.coli*, *enterococci*, and coliphage (MS-2)). Samples were collected from July of 2000 through July of 2001. Some months it was impossible to obtain a sample from every well site. Obstacles to obtaining water samples at each site every month included frozen faucets, mail delays, laboratory accidents, and homeowners being unavailable when they were contacted for sampling. Replacement samples were collected as soon as possible if these events occurred at a sampling site. A minimum of two liters of unchlorinated source water was collected

from each sampling site on each sample date. Water samples were shipped to the SLOH via United Parcel Service for laboratory analysis.

E.Coli And Enterococci

E.coli and the *enterococci* were detected using the Colilert™ and Enterolert™ methods.

Colilert uses 2 nutrient-indicators, ONPG and MUG. The nutrients are the major sources of carbon in Colilert and can be metabolized by the *coliform* enzyme β -galactosidase and the *E. coli* enzyme β -glucuronidase. As *coliforms* grow in Colilert, β -galactosidase is used to metabolize ONPG and changes the samples color from clear to yellow. The yellow color indicates *coliform* are present. *E.coli* uses β -glucuronidase to metabolize MUG and create fluorescence. Fluorescence indicates the presence of *E. coli* (IDEXX, 1990-1999). The Colilert test provided both *coliform* and *E.coli* results.

Enterolert uses a Defined Substrate Technology® (DST®) nutrient-indicator to detect *enterococci*. This nutrient-indicator fluoresces when metabolized by *enterococci* (IDEXX, 1990-1999).

Quantitative results for both Colilert and Enterolert were obtained by separating 100 mL of the water sample with the respective nutrient base added into heat-sealed Quanti-trays²⁰⁰⁰ and inspecting cells after 24 hour incubations at 35°C.

Coliphage

Two hundred and fifty milliliter samples were utilized for coliphage analyses. An enrichment technique approved in November of 1999 by the EPA for groundwater monitoring was used (EPA Method 1601) (Battigelli 1999). Each sample was supplemented with nutrients to support bacterial growth. Bacterial host cells specific for each of two types of coliphages: (1) those which infect bacteria via adsorption to external physical appendages used for sexual conjugation (male-specific coliphages), and (2) those which infect bacteria via direct adsorption to the cell wall (somatic coliphages) were added to the samples. The samples were incubated overnight at 37°C to allow bacterial growth. Bacterial viruses (coliphages) present in a sample will grow due to the abundance of host bacteria. Droplets from the enrichment cultures were spotted onto fresh lawns of bacteria. Samples positive for coliphages were identified according to zones of lysis which develop on inoculated lawns within hours of inoculation.

Gene Probe Detection Of Pathogens In Phase II Of Illness Related Water Samples

A cooperative agreement was set up with North Shore Clinic (Door County Memorial Hospital) to recruit gastrointestinal illness volunteers. Patients diagnosed by medical staff at North Shore Medical Clinic with gastrointestinal illness based on clinical symptoms were provided information on the drinking water study. Individuals who agreed to participate in the study were surveyed to identify demographic information, their main drinking water source, potential transmission by person-to-person contacts, recent history of gastrointestinal symptoms, and other potential sources of their illness. WI DNR staff contacted participants referred from the hospital to schedule a time to

collect water samples from the patients primary water source. Water samples were analyzed by the SLOH for *coliform*, and fecal indicators (*E.coli*, *enterococci*, and coliphage).

Gene probe environmental pathogen tests were conducted on wells that had multiple positive results reported for fecal indicator tests or waterborne illness was strongly suspected based on the patient's symptoms. Stool samples of ill individuals were also collected when available. The stool samples were tested for cultures of *salmonella*, *shigella*, *campylobacter*, *E.coli* 0157:H7, *yersinia*, *vibrio*, *aeromonas*, *pleisiomonas*, and *edwardsiella*.

A large volume water sample was collected according to the method described by the EPA's Information Collection rule (Battigelli 1999). A novel gene probe method was substituted for the cell culture for the diagnostic assay portion of the procedure. This method has already been used by the SLOH for over four hundred groundwater isolates collected from three states in order to characterize the extent of viral pollution in different hydro-geological settings. Large volumes of groundwater (200-400 gallons) were filtered on-site through positively charged filters in order to capture virus particles. Viruses are eluted into small volumes at the analytic laboratory using an alkaline protein solution (1.5% beef extract/0.05 M glycine, pH 9.5) and concentrated by organic flocculation. Following centrifugation, virus concentrates are supplemented with lysis buffer (6M guanidinium hydrochloride) and subjected to acidic phenol-chloroform extraction to recover viral ribonucleic acid (RNA). RNA are then reverse-transcribed into cDNA,

amplified enzymatically according to the polymerase chain reaction™ and visualized in 2.0% agarose electrophoretic gels. DNA is then recovered from the gels onto nylon membranes and hybridized to nonisotopic probes specific for each virus group/family (Battigelli 1999). The virus groups/families to be tested for included astrovirus, caliciviridae (ex. calicivirus, hepatitis E, Norwalk and Norwalk like viruses), enterovirus (ex. coxsacki, hepatitis A, and polio,) and rotavirus.

Data Analysis

Statistical analysis was completed utilizing SAS software. The four different microbiologic test method results were used to rank the 25 well sampling sites. Ranks for the 25 well sites were calculated for each of the four indicators in SAS. The total rank for each site was obtained by summing the four site-specific ranks and then ranking the 25 rank sums in SAS. The total ranks were used to determine the best and worst water quality wells and discuss possible reasons for differences in the sample results.

The average monthly temperature and precipitation data were graphed with counts of the *coliform* detects for each month of the study to attempt to identify a relationship between the data. The precipitation and temperature data were collected for Door County at the Wisconsin Extension Peninsula Agricultural Research Station which is centrally located within the county.

The four microbiologic indicators were analyzed with Pearson's Correlation in SAS to determine similarities and differences between the methods. The Pearson's Correlation

Coefficient was utilized to make a determination of what variables may be statistically significant in explaining when *coliform* contamination would occur. Regression models for the parameters that have significant Pearson correlation with *coliform* were generated. The logarithmic transforms for *coliform* counts were used as the dependent variables to normalize the distribution of the data. Temperature and precipitation values were used as independent variables. Other independent variables that were analyzed include year of the well construction, casing depth and well depth. The greater prevalence of *coliform* detects in the samples provided more data than the other indicators in this study to use for modeling.

CHAPTER 4 RESULTS AND DISCUSSION

Results Of 25 Wells Based On Rank

The sample results from the 25 well sites tested monthly for the four indicator tests were compared. One tool used to look at the data was ranking the well sites for each indicator and then calculating a total rank based on all four indicators. *Enterococci* and coliphage provide some additional insight in classifying wells that the traditional *coliform/E.coli* testing would miss. This ranking is useful in comparing the differences in the best water quality wells and worst water quality wells with soil, karst, and casing requirement information (Figure 2.1, Figure 2.2, & Figure 4.6). The 25 well sites were ranked based on the positive detects in the four indicator tests with equal weighting on each test. The total rank assigned to each well ranged from 1 to 25 in order of best to worst quality water (Table 4.1).

The five best water quality wells (sites # 1, 3, 4, 10, & 21) were not located near active farming and constructed in 1986 or later. The five best water quality sites averaged less than three karst features identified within ½ mile. The average for the five wells sites was 1.8 karst features.

Four of the five worst water quality wells (sites # 5, 7, 18, & 23) were constructed prior to 1986. One of the six wells (# 15) was a replacement well constructed in 1986 in a known aquifer contamination area. Three out of the five wells (sites # 7, 15, & 23) with the worst quality water were located near active farming operations. Two of the wells

(sites # 7 & 15) near farm sites also had more than three karst features within ½ mile of the well¹. Well # 18 had five karst features within ½ mile. Well # 5 has three karst features identified within ½ mile of the site. The average number of karst features for the four Northern Door sites is 4.5.

Table 4.1: Water Quality Rank Of 25 Well Sites

(Coliform, *E.coli*, *Enterococci*, and Coliphage Ranks are based on 25 being the best quality well and 1 being the worst with the most indicator detects. The total rank is based on each indicator weighted equally by summing the individual indicator ranks and ranking the sum).

BEST WATER QUALITY WELLS

WORST WATER QUALITY WELLS

Map #	Coliform Detects	Coliform Rank	<i>E.coli</i> Detects	<i>E.coli</i> Rank	<i>Enteroc.</i> Detects	<i>Enteroc.</i> Rank	Coliphage Detects	Coliphage Rank	Ranks Sum	Total Rank
1	0	4	0	9.5	0	8	0	9.5	31	3
2	2	9.5	0	9.5	0	8	0	9.5	36.5	6.5
3	0	4	0	9.5	0	8	0	9.5	31	3
4	0	4	0	9.5	0	8	0	9.5	31	3
5	8	22.5	4	23.5	4	22.5	0	9.5	78	23
6	0	4	0	9.5	0	8	1	22	43.5	11.5
7	8	22.5	2	21.5	4	22.5	1	22	88.5	24
8	4	15.5	0	9.5	0	8	1	22	55	18
9	5	17.5	0	9.5	1	17.5	0	9.5	54	16.5
10	0	4	0	9.5	0	8	0	9.5	31	3
11	0	4	0	9.5	0	8	1	22	43.5	11.5
12	3	12.5	0	9.5	1	17.5	0	9.5	49	14
13	3	12.5	0	9.5	0	8	0	9.5	39.5	8.5
14	3	12.5	0	9.5	0	8	0	9.5	39.5	8.5
15	11	24.5	6	25	7	25	1	22	96.5	25
16	11	24.5	1	19.5	1	17.5	0	9.5	71	19
17	1	8	0	9.5	0	8	1	22	47.5	13
18	6	20	4	23.5	2	20	0	9.5	73	21
19	6	20	1	19.5	4	22.5	0	9.5	71.5	20
20	2	9.5	0	9.5	0	8	0	9.5	36.5	6.5
21	0	4	0	9.5	0	8	0	9.5	31	3
22	5	17.5	0	9.5	1	17.5	0	9.5	54	16.5
23	6	20	2	21.5	4	22.5	0	9.5	73.5	22
24	3	12.5	0	9.5	0	8	1	22	52	15
25	4	15.5	0	9.5	0	8	0	9.5	42.5	10

¹Well site 23 may have karst features. The karst features in the area around site # 23 in Southern Door County have not been mapped.

Analysis Of Indicator Tests

The Pearson's Correlation Coefficient indicated that there were positive relationships between *coliform*, *enterococci*, and *E.coli* (Table 4.2).

Table 4.2: Pearson Correlation Matrix

	<i>Coliform</i>	<i>E.coli</i>	<i>Enterococci</i>	Coliphage
<i>Coliform</i>		0.46935 Prob. < 0.0001	0.66454 Prob. < 0.0001	0.08037 Prob. < 0.1700
<i>E.coli</i>	0.46935 Prob. < 0.0001		0.72531 Prob. < 0.0001	-0.00825 Prob. = 0.8884
<i>Enterococci</i>	0.66454 Prob. < 0.0001	0.72531 Prob. < 0.0001		0.00335 Prob. = 0.9546
Coliphage	0.08037 Prob. < 0.1700	-0.00825 Prob. = 0.8884	0.00335 Prob. = 0.9546	

The *coliform* relationship with the bacteriological fecal indicators was not random. There was a moderate positive association of the occurrence of *enterococci* and *E.coli* that was statistically significant. The correlation values were squared to assess the explanatory power of each of the bacterial indicators on the other. The *E.coli* explained 22% of the variance in *coliform* detects. The *enterococci* explained 44% of the variance in *coliform* detects. The greater explanatory power of *enterococci* suggests it may be an asset for identifying water supplies contaminated with fecal material.

The relationship between the bacteriological fecal indicators was also not random. The variables explained 53% of the variance in the other indicator and the relationship was significant. The higher correlation values were expected because both indicators originate from fecal material and *coliform* bacteria have many other sources.

The viral coliphage indicator did not correlate with any of the other indicators according to the Pearson Correlation analysis, suggesting it was a random relationship. Based on the fact that *E.coli* is the food source for these viruses there should be some relationship but other factors not considered in this analysis may play a role in why *E.coli* was not significantly related to coliphage. Viruses generally have longer survival times than bacteria. Viruses are also smaller in size than bacteria. These factors may allow viruses to exist in the environment longer and travel further. Therefore, a water sample may not contain *E.coli* and coliphage at the same time because coliphage could have been transported from a greater distance or been a remnant of a prior *E.coli* contamination event in the local aquifer.

Results Of Indicator Tests Per Month For All Wells

Precipitation events and temperature are believed to provide some explanation of the frequency and degree of groundwater contamination by microbes. Precipitation and temperature data were obtained from the Door County Wisconsin Agricultural Peninsular Research Station that is centrally located in the county.

The average monthly rainfall and snowfall for 1970 – 2000 is listed in the following table along with the values the research station recorded for these events in 1999-2000 (Table 4.3).

Table 4.3: Peninsular Research Station Precipitation Data (Peninsular 2000-2001)

	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
Historical Rainfall	1.79	1.11	2.12	2.67	2.92	3.49	3.41	3.61	3.43	2.69	2.53	1.76
Rainfall During Study	1.60	1.80	0.50	3.75	5.37	3.50	0.29	4.75	4.40	0.96	3.27	2.09
Historical Snowfall	15.7	8.2	7.7	2.2	0.1	0.0	0.0	0.0	0.0	0.1	3.3	11.6
Snowfall During Study	3.0	7.0	4.5	3.0	0.0	0.0	0.0	0.0	0.0	0.0	8.0	25.0

(monthly precipitation reported in inches)

* Historical data collected from 1970 – 2000

Low Precipitation Compared To Historical Average

High Precipitation Compared To Historical Average

The precipitation events for March, July, and October were abnormally low during this study period compared to the historical average. The lack of precipitation may partially explain why the individual counts of indicator bacteria rapidly decreased in October samples compared to August and September water samples. Precipitation can carry surface contaminants with it as it seeps into the ground or bedrock. The July counts of indicator bacteria were also low in comparison to the higher rain month of August. However, the differences in contamination concentrations may be due to the colder temperature differences encountered in October and the impact it has on diminishing microbes in the environment. The fecal indicators were highest in August and September. The rainfall was also higher during these months. Conversely, the 2001 snowfall accumulations were low compared to the historical averages. The early end to winter in 2001 did not yield the same amount of runoff or melt water to impact the groundwater as an average year would have yielded.

These responses can be seen in the following graph that shows the percentage of microbe indicator detects compared with each month's average precipitation (Figure 4.1).

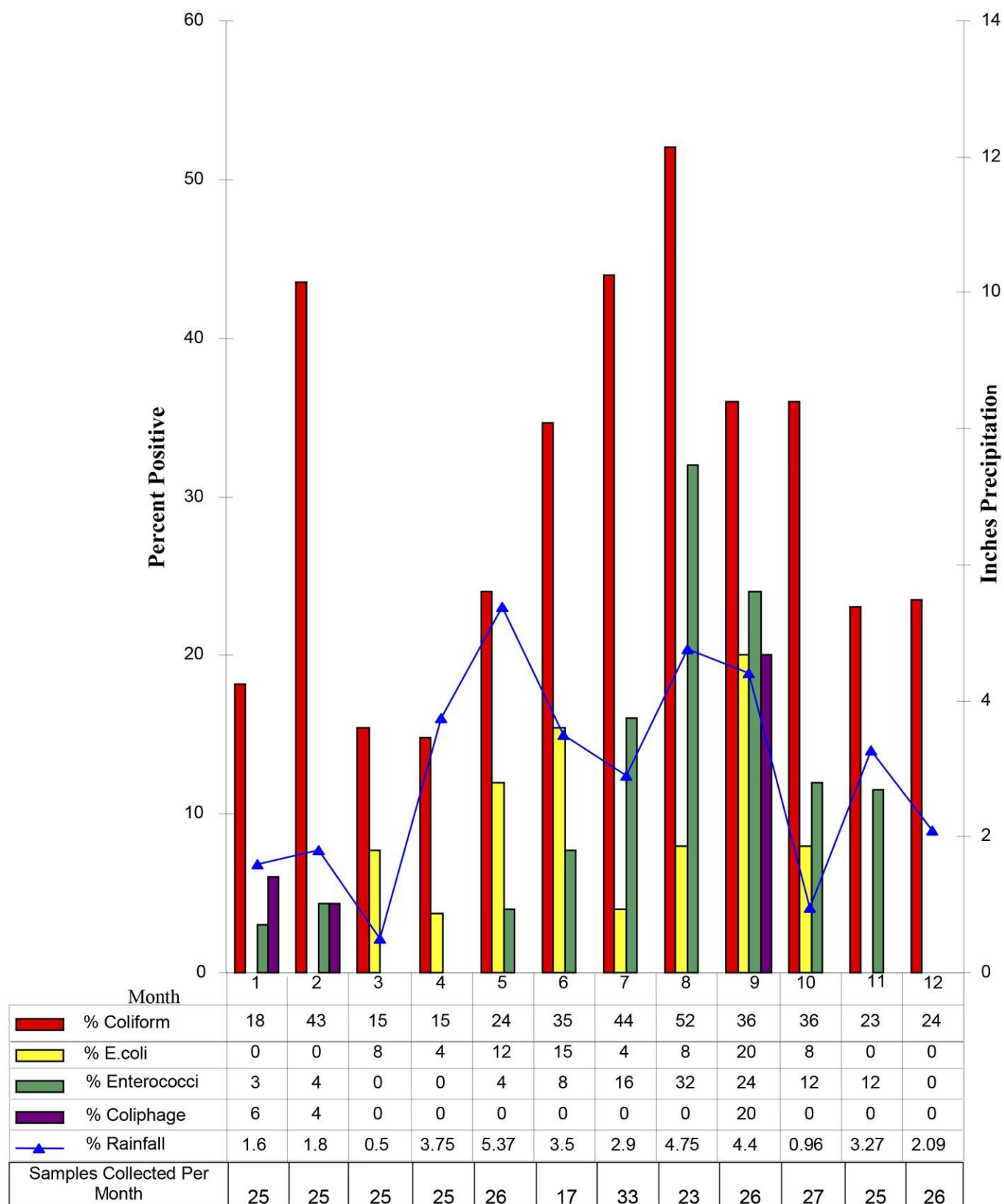


Figure 4.1: Percent Of Positive Indicators And Rainfall Per Month

Temperature is also thought to play a part in the occurrence of detects of microbes in drinking water systems. The following table shows data from the Peninsular Research Station for the monthly historical average temperature and the temperature recorded for each month during this study (Table 4.4).

Table 4.4: Peninsular Research Station Temperature Data (Peninsular 2000-2001)

	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
Historical High Temperature *	24.3	27.9	37.8	50.4	63.9	73.5	78.5	76.5	68.6	56.0	41.7	29.5
High Temperature During The Study	29.5	27.9	37.7	55.4	65.2	71.9	75.5	76.2	67.7	59.5	42.4	23.4
Historical Low Temperature *	7.8	10.9	21.4	32.4	42.8	52.6	58.6	57.4	49.9	38.8	27.8	16.0
Low Temperature During The Study	15.4	11.5	21.8	36.3	44.9	54.0	57.9	58.1	49.8	43.1	31.5	8.2

(Temperatures reported in degrees Farenheight)

* Historical data collected from 1970 – 2000

Low Temperature Compared To Average

High Temperature Compared To Average

The historical averages for recorded monthly temperature data collected from 1971 through 2000 suggest that the monthly temperatures during this study were higher than normal in the months of January, April, and October. The temperature in the month of December was significantly lower than normal. These differences in temperature may have led to less positive indicator results in the month of December but more positive indicator results in the months of January, and April during the study. An increase in October positive indicator samples may have been offset by the less than average rainfall in 2000. However, the percentage of *coliform* detects in this study do stay constant in September and October (Figure 4.1). Normally, a decrease with the decreasing temperature would be expected.

The average air temperature for each month appears to have some relationship with the percentage of positive microbe indicators from the well water samples (Figure 4.2). The month of February does not appear to follow the general trend shown in the graph.

However, the periods of warmer weather during January and February combined with rainfall may have allowed snow to melt and the ground to thaw and allow contaminated water to seep into the aquifer.

The strongest relationship between temperature and an indicator is shown with *coliform* bacteria. This is likely due to the greater prevalence of *coliform* in the environment. The *enterococci* appear to be following a similar trend to *coliform* but on a smaller scale. The *E.coli*. may be less resilient in the environment and show up when recent fecal sources are released and rainfall washes them into the aquifer. The coliphage detects do not appear to follow the changes in temperature.

The previous responses discussed can be seen in the following graph of indicator detects per month compared with temperature data (Figure 4.2).

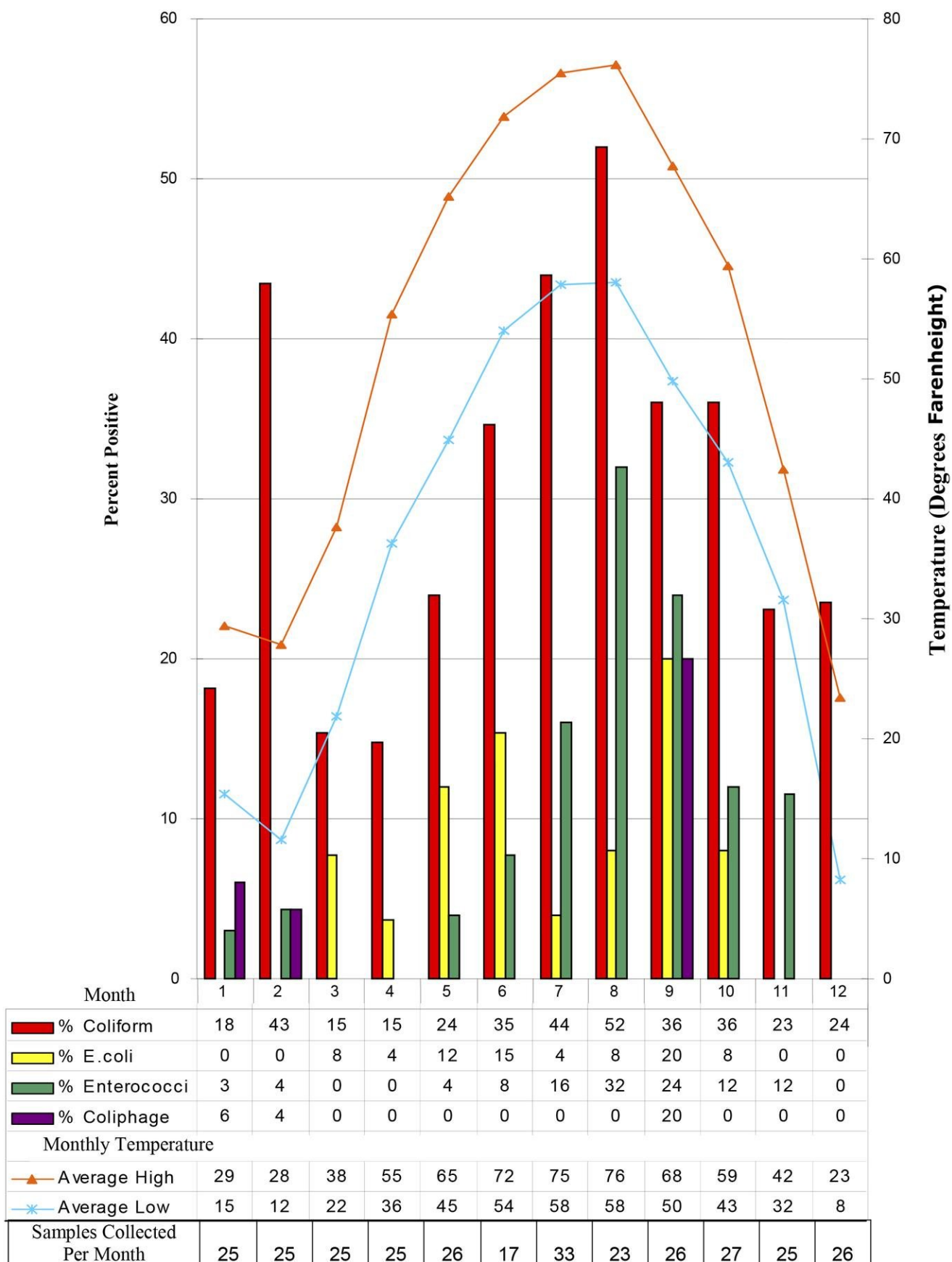
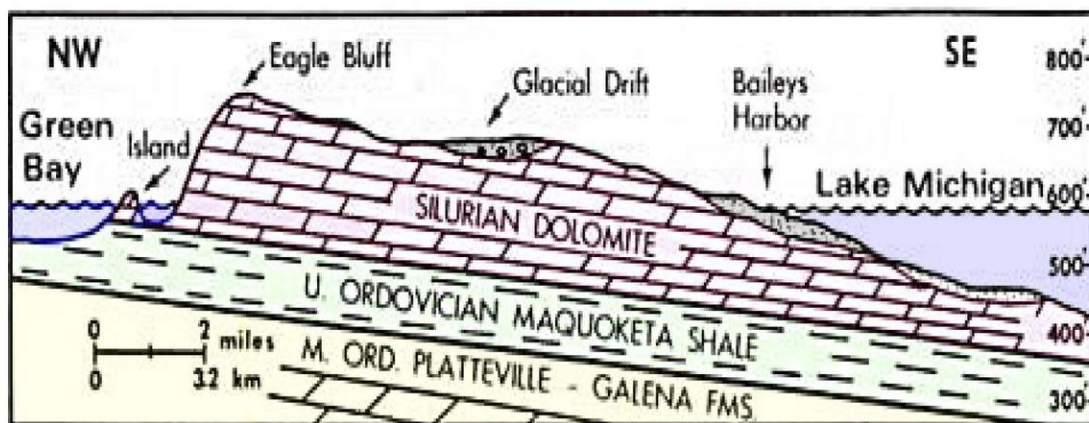


Figure 4.2: Percent Of Positive Indicators And Rainfall Per Month

Least Squares Regression Of Well *Coliform* Sampling With Environmental Factors

A least square regression model was fit to compare what factors at the 25 well sites were significant when compared with the log of *coliform* water results. A prior study of 192 wells in Door County failed to find a significant relationship between well casing depth and *coliform* or depth of a well and *coliform* (Hutchinson 1992). A decrease in *coliform* contamination would be expected for newer wells due to changes in the well construction requirements to provide greater protection for wells from microbial contamination. The significance of the amount and when precipitation occurred prior to sampling of the well was also expected to show some relevance to the amount of *coliform*.

The data collected from well site # 19 in this study were not utilized in the regression modeling. The reason for this elimination is that site 19 has a well that is constructed through Ordovician Maquoketa Shale (Figure 4.3). The shale acts as a confining layer or barrier to prevent water in the upper Niagara dolomite aquifer and the lower Galena sandstone/dolomite aquifer from mixing. Therefore, the water in the lower aquifer is not as susceptible to surface contamination sources. Well # 19 is 640 feet in depth. This is more than double the depth of any other well included in the study. The well was expected to be a control site, with no bacteriological contamination. However, the well had an overlapping well cap at the start of the study that allowed earwigs to enter the well. The combination of the greater depth, shale confining layer and the source of the bacterial contamination in the well make the data unrepresentative of the other wells.



Adapted From Paull, R.K., R.A. Paull 1988

Increasing Depth ↓	Rock Unit	Aquifer	Formation
	Silurian Dolomite	Niagaran Series Dolomite	Engadine
			Manistique
			Hendricks
			Byron
		Alexandrian Series Dolomite	Mayville
	Ordovician Maquoketa Shale	Confining Bed	Maquoketa
	Ordovician Dolomite/Sandstone	Upper Sandstone	Galena

Adapted From Sherrill 1978

Figure 4.3: Door County Geologic Cross Section And Stratigraphy

Well site 19 would not likely show responses to surface contamination events or temperature changes. The following graph shows well Map # 19 as an outlier compared to the other wells included in the study (Figure 4.4).

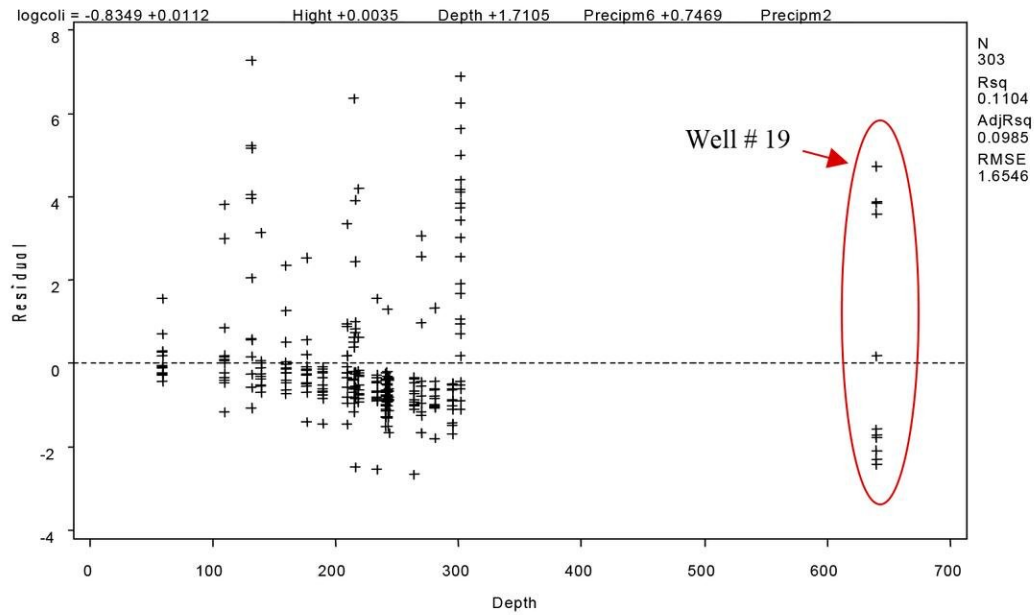


Figure 4.4: Plot Of Regression Residuals Verses Depth

The graph shows the depth of well # 19 is more than double any other well included in the study. The following graph shows the well site data without well # 19 (Figure 4.5).

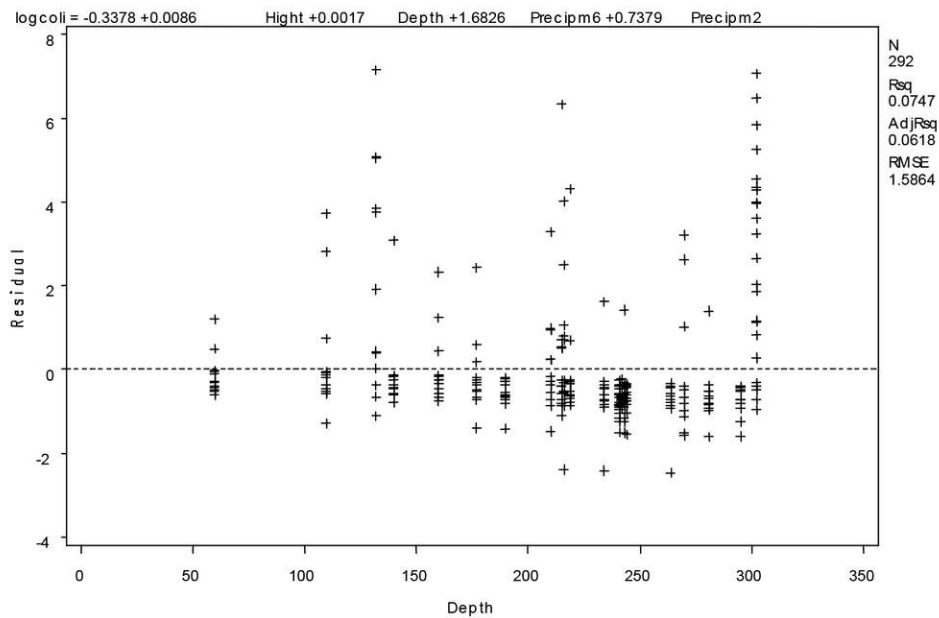


Figure 4.5: Plot Of Regression Residuals Verses Depth Without Well # 19

Regression models were fit with the logarithmic value of the *coliform* count as the dependent variable and independent variables from weather information and data on the 24 wells. The initial independent variables were selected based on Pearson Correlation values significant at the 0.05 level. All precipitation data and total depth were also included because these factors are similar to variables that were found significant in the Pearson Correlation values (Table 4.5).

Table 4.5: Independent Variables Used In Model

Casing Depth	Total Depth	High Temperature	Low Temperature	Year of well construction	Precipitation – (day of sampling and 1 – 8 days lag)
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The regression model was first fit with the yearlong 2000-2001 data set (Table 4.6).

Table 4.6: Least Squares Regression Model With Year Round Precipitation Data

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_4 X_4 + \text{error}$$

Terms

Y = log of coliform (dependent variable)

b₀ = y intercept

b₁ = casing parameter estimate

b₂ = high temperature parameter estimate

b₃ = precipitation 6 days lag parameter estimate

b₄ = year well constructed parameter estimate

X₁ = casing depth

X₂ = high temperature

X₃ = precipitation 6 days lag

X₄ = year well constructed

The model is significant based on the low probability (0.0001) that the independent variables had no impact on the dependent variable (Table 4.7). The F value of 15.20 indicates that collectively the independent variables in the model had a limited linear relationship with the dependent variable. The independent variables collectively explained 18% of the variability in the *coliform* counts.

Table 4.7: Regression Model Analysis Of Variance With Winter Data

F Value = 15.20	Probability > F = 0.0001	R-square = 0.1810
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The terms that were determined to be significant in the model are listed in Table 4.8.

Table 4.8: Regression Parameter Estimates With Winter Data

Variable	Parameter Estimate	Standard Error	F Value	Probability (> F)
Casing Depth	0.01532	0.00274	31.15	0.0001
High Temperature	0.01015	0.00463	4.79	0.0294
Precipitation 6 days lag	1.41566	0.42884	10.90	0.0011
Year Well Constructed	-0.04938	0.00828	35.61	0.0001

The year of well construction had a negative relationship with the log of the *coliform* count data¹. This suggests that older wells are more likely to be contaminated. This is physically sensible since older wells may not have been grouted as effectively as drillers now grout newer wells. Overlapping caps installed on older wells may also provide some explanation for the greater occurrence of *coliform*. The natural degradation of the well casing and grout over time is another possible explanation for the observed relationship. Older wells may also have less casing due to changes in the well construction requirements for Door County that occurred in 1957 and 1972. However, the casing depth variable in the regression model was a significant term with a positive relationship, increasing along with *coliform* detects. One possible explanation for this inconsistency is that the preset casing requirements are based on geologically sensitive areas verses those areas with greater natural protection. The casing depth is determined in Door County by

¹*Coliform* counts are a statistical number for the colilert test based on the historical method of membrane filter testing which provided the number of colonies that grew on the filter. The number provides a magnitude to use as a reference for the severity of contamination.

location. Zones of 100 feet and 170 feet were established in the 1970s for drilled wells constructed in the fractured dolomite formation.

Examples of areas believed to provide greater protection from surface contamination are: wells drilled into the Alexandrian Series of dolomite, areas with small water level fluctuations and locations where there is more unconsolidated material. The less vulnerable areas only require 100 feet of casing. Greater casing is required in areas that are suspected of being more vulnerable to surface contamination due to less soil, areas with large water level fluctuations, and/or drilled into the Niagaran Series of the dolomite (Figure 4.3).

The more vulnerable areas require 170 feet of casing. Therefore, wells constructed within areas with the greater casing requirements are in areas that are also more likely to have surface contamination impacting the wells. This study did not show that greater casing depth yielded less microbiologic contamination coincident with the zoned casing requirements.

Wells of varying casing depth would need to be constructed within areas containing similar geologic formation and soil depth to determine the impact the deeper casing has on contamination. The sites in this study did not provide homogeneous geologic settings to compare the impact of increasing casing depth. Figure 4.6 shows the wide variety of the well sites in relation to the minimum casing requirements included in this study. The daily high atmospheric temperature also showed some significance with the log of the *coliform* counts in the regression with the entire years worth of data. As the temperature

increased the *coliform* count also increased.

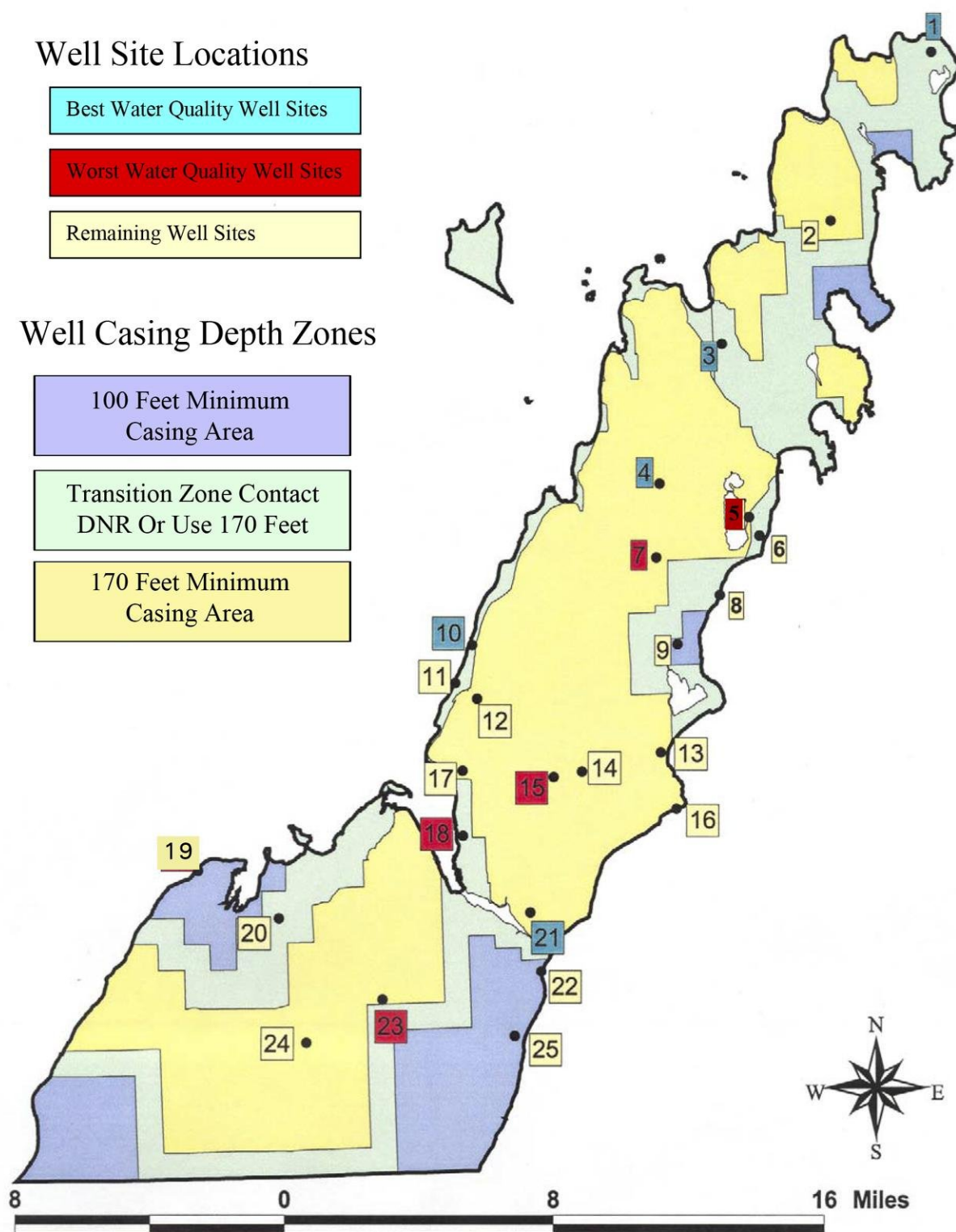


Figure 4.6: Door County Well Casing Requirements With Study Sites

The units in the model were standardized in SAS to compare the relative strength of the independent variables on the logarithmic *coliform* count. The year the well was constructed had the most influence. The logarithmic value of *coliform* would decrease 45% standard deviation for each standard deviation increase in the year the well was constructed, if the other variables remained constant (Table 4.9). The casing depth was also shown to have a strong influence. If the other variables remained constant the logarithmic *coliform* counts would yield a 42% standard deviation increase for each standard deviation of increase in casing depth. The variables high temperature and precipitation 6 days lag were considerably weaker predictors of the response in *coliform* counts.

Table 4.9: Comparison Of Variables In Standardized Units

Variable	Standardized Estimate
Casing Depth	0.42186
High Temperature	0.12049
Precipitation 6 days lag	0.18219
Year Well Constructed	-0.45025

The model was also fit with the winter month data removed (Table 4.10) due to concerns that the snow melt and precipitation events would not be accurately accounted for in the model. This is because the predicted amount of melt water infiltrating an area couldn't be determined with the data available.

Table 4.10: Least Squares Regression Model With Winter Data Removed

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + \text{error}$$

Terms

Y = log of coliform (dependent variable)

b0 = y intercept

b1 = casing parameter estimate

X1 = casing depth

b2 = precipitation 1 day lag parameter estimate

X2 = precipitation 1 day lag

b3 = year well constructed parameter estimate

X3 = year well constructed

The model is significant based on the low probability (0.0001) that the independent variables had no impact on the dependent variable (Table 4.11). The F value of 12.78 indicates that collectively the independent variables in the model had a limited linear relationship with the dependent variable. The F value decreased slightly in the model fit excluding the winter data compared to the model fit with winter data included. The independent variables collectively explained 19% of the variability in the *coliform* counts. This was a slight increase compared to the model fit with the winter data.

Table 4.11: Regression Model Analysis Of Variance With Winter Data Removed

F Value = 12.78	Probability > F = 0.0001	R-square = 0.1943
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The significant variables did change with the months of December, January, February, and March removed from the analysis (Table 4.12).

Table 4.12: Regression Parameter Estimates With Winter Data Removed

Variable	Parameter Estimate	Standard Error	F Value	Probability (> F)
Casing Depth	0.01833	0.00392	21.89	0.0001
Precipitation 1 day lag	0.75656	0.23560	10.31	0.0016
Year Well Constructed	-0.05366	0.01188	20.40	0.0001

During this data fit the precipitation 1-day lag to sample collection was found to be the most significant with occurrence of *coliform*. The year of well construction and log of the *coliform* concentration still had a significant negative relationship in this model. As the age of a well increased so did the *coliform* contamination. The *coliform* appeared to increase as casing depth increased based on the significant positive relationship between the two variables. The precipitation 1-day lag was more significant with the winter month data removed from the model than it was with the entire year of data in the model. The daily precipitation data in both models for one year is not adequate to determine an average breakthrough time for its impact on the aquifer. Data over an extended period would be better able to narrow the amount of precipitation needed to impact the average Door County well. Use of public water sampling data may be useful in future work to obtain this estimate.

Another regression model was fit with the precipitation data from the sample date and 8 days lag summed into one term - total rainfall (Table 4.13). The entire year of study data was utilized in the model.

Table 4.13: Least Squares Regression Model With Winter Total Rainfall Term

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + \text{error}$$

Terms

Y = log of coliform (dependent variable)

b0 = y intercept

b1 = casing parameter estimate

X1 = casing depth

b2 = total rainfall 8 days lag parameter estimate

X2 = total rainfall 8 days lag

b3 = year well constructed parameter estimate

X3 = year well constructed

The total rainfall model is significant based on the low probability (0.0001) that the independent variables had no impact on the dependent variable (Table 4.14). The F value of 17.23 indicates that collectively the independent variables in the model had a limited linear relationship with the dependent variable. The F value was the highest out of the three models suggesting it was the most linear relationship. The independent variables collectively explained 16% of the variability in the *coliform* counts based on the R-Squared value. Therefore, this model provided a slightly lower level of explanation for the variability in the *coliform* counts.

Table 4.14: Regression Model Analysis Of Variance With Winter Data Removed

F Value = 17.23	Probability > F = 0.0001	R-square = 0.1578
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The significant variables included casing depth, total rainfall for 8 days lag, and year of well construction (Table 4.15).

Table 4.15: Regression Parameter Estimates With Rainfall Data Summed

Variable	Parameter Estimate	Standard Error	F Value	Probability (> F)
Casing Depth	0.01543	0.00278	30.84	0.0001
Total Rainfall 8 days lag	0.27823	0.08895	9.78	0.0019
Year Well Constructed	-0.05002	0.00837	35.70	0.0001

The temperature variable that was significant in the earlier year-round regression model is not significant in the model with a total rainfall amount for the eight days lag. Overall the modeling suggests that the rainfall, and the year of well construction were significant factors in explaining water quality.

***Coliform* Count Relationship With Precipitation**

The well *coliform* indicator counts were plotted by sample date with the rain events the day of sampling and 8 days lag to visualize if the model results were correct by predicts that rainfall events were influencing a particular wells water sample result. The symbols for the *coliform* concentration and rain events are listed in the tables below (Table 4.16).

Table 4.16: Precipitation And *Coliform* Graph Symbol Key

Count of <i>Coliform</i> In Water Sample					Graph Symbol				
					*				
Precipitation Event	Day of Sampling	1 day lag	2 days lag	3 days lag	4 days lag	5 days lag	6 days lag	7 days lag	8 days lag
Graph Symbol	0	1	2	3	4	5	6	7	8

The well # 15 graph does suggest the well is influenced by precipitation events (Figure 4.7).

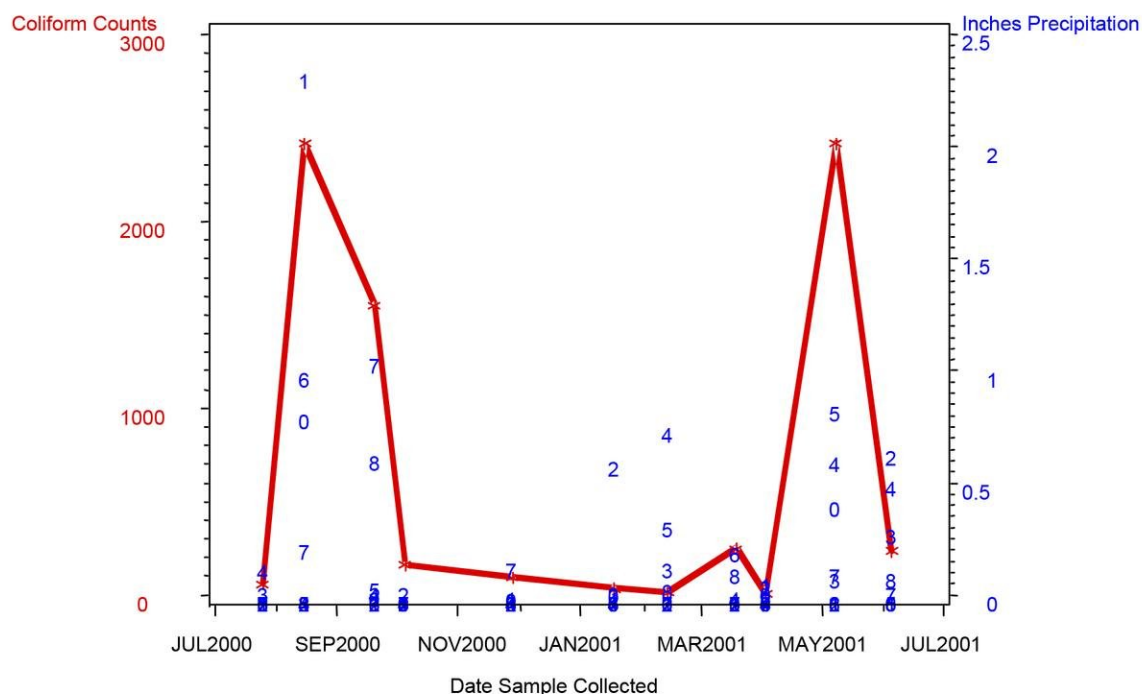


Figure 4.7: Well # 15 *Coliform* And Precipitation Data Verses Date Sample Collected

Well site # 15 provides the best visual of precipitation events having an impact on the *coliform* counts in the water samples. Graphs of other wells in the study also show some response that may be explained in part by precipitation events. Graphs of individual sites are located in Appendix B.

Soil and karst features may explain some of the variability in water quality among well sites. Karst feature surveys were not done for each individual site in this study. The karst information used to create the maps for this study are from a geographic information layers that includes data of known features from prior studies. For example, the layer data does contain sink holes and fractures that are known to be located on the

property near well site # 15. However, Figure 2.2 does show many karst features within a few miles of site # 15. The karst information layer does help identify areas of concern but it is not inclusive enough to rely on to exclude karst features as a possible source of contamination because many features are not included in the data layer. The karst information layer was overlaid on aerial photos along with soil information and is included in Appendix A. Figure 4.8 shows the well site # 15 and its proximity to shallow soils and karst features that have been identified in previous studies (Stieglitz 1986, USDA 1978, USGS 1992).

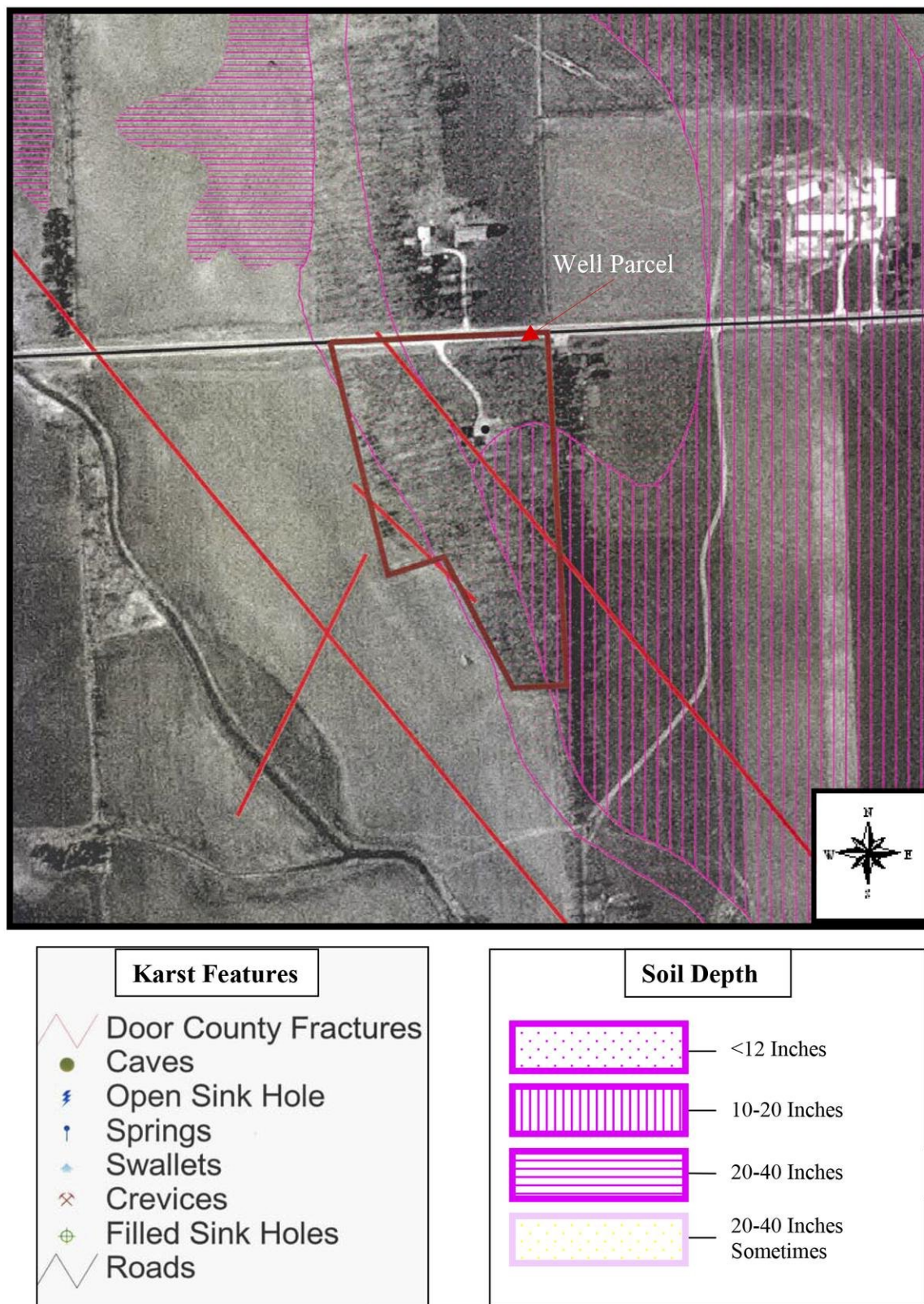


Figure 4.8: Well Site # 15 Air Photo With Shallow Soil And Karst Information

Results Of Illness Testing

The illness portion of this study was finalized with the North Shore Medical Clinic in March 2001. This portion of the study was plagued with problems. The protocol for stool and water was finalized in October 2000, but the hospital was unable to begin referring patients until March 2001. The major spring melt had already occurred prior to the hospital activation of sampling protocol. Earlier hospital staff pediatrician involvement would also have been desired since numerous stool samples were collected before the hospital had notified their staff of the referral process. The hospital referred one illness case during the study. Ideally a referral process in place in the fall would have provided the best time to catch virus detects based on the seasonal patterns of viruses identified in other virus studies. State Laboratory of Hygiene staff changes and lack of familiarity with the testing methods also led to incomplete viral testing of the water samples and stool samples that were submitted in the spring 2001. Due to the cost of the tests in this study, few samples were collected. Unfortunately no incidence could be confirmed where pathogen detects in both the well water and a water consumer's stool sample were identified to contain the same pathogen during this study.

There were five well sites that were tested for specific viruses based on positive fecal indicator tests or based on illness symptoms of individuals consuming the water. The first sample was collected in October 2000. Stool samples were also collected from two members living in the residence. Unfortunately, for the study the residents did not contact the study when the diarrhea symptoms began and would have yielded the highest concentrations of pathogens in the stools. The family doctor had diagnosed two members

in the family with waterborne disease based on their symptoms. However, the testing kit for stool samples and process to preserve them was not finalized when this event occurred. Therefore, as an alternative, the local health departments stool kits were used for sample collection. These kits used preservatives and a collection method that was not well suited to later run RT-PCR testing¹.

Routine cultures for the 2 stool samples were negative for Salmonella, Shigella, Campylobacter, Escherichia coli 0157, Yersinia, Vibrio, Aeromonas, Plesiiomonas and Edwardsiella. The well water sample results from the residence indicated *coliform* contamination and also *enterococci*. Additional, viral testing of the well water with RT-PCR could not be scheduled because the well was shut down for the season prior to finalizing the water collection procedure with the lab. The well consisted of an old 4-inch diameter casing. Based on the casing diameter it is likely that the well was shallow but no data was available on the well construction. The property was on the shore of Green Bay. The dolomite bedrock escarpment was layered with homes upgradient of the well. It is likely that the groundwater from failing septic systems would discharge towards the bay. Fluids from the neighbors septic systems could pass through the property and have the potential to contaminate the well.

There were three stool samples and four water samples submitted in spring of 2001 for testing. Three stool samples were collected from ill individuals that agreed to participate

¹RT-PCR is the gene probe process described in the methods section where viral RNA is used to reverse transcribe DNA. Copies of the DNA sequence are created to identify the organism.

in the study. Two of the ill individuals were consuming water from the same well. The stool samples were not analyzed due to changes at the lab.

Two of the four water samples submitted in the spring of 2001 were collected from the homes of the ill individuals that submitted stool samples. In addition to these two samples, two water samples were taken from the 2 most contaminated wells out of the 25 wells in the monthly monitoring portion of this study. All four of the water samples were not found to contain enterovirus, hepatitis A, or rotavirus. Due to changes in lab personnel the controls could not be found to do the testing RT_PCR for calicivirus and Norwalk viruses.

The linkage of human illness to groundwater is a suggested problem based on the contacts the DNR receives from people who become ill with gastrointestinal symptoms shortly after returning to Door County (WI DNR¹ Unpublished Door County Files). Funding to investigate these complaints and provide the testing is not readily available. This is an area of study where additional work would be beneficial.

CHAPTER 5 CONCLUSIONS AND RECOMMENDATIONS

This study provided a preview of the percentage of EPA proposed fecal indicator tests that may yield positive detects in Door County public TN systems. As was expected, the data show that there is some vulnerability for Door County wells to microbial contamination. For example, half of the wells did have *coliform* detected in water samples collected in August.

However, more positively the additional tests proposed in the GWR did not drastically increase the number of unsafe wells. Prior coliphage indicator testing in Wisconsin had suggested 95% of the wells would be impacted (Janczy 1998). Only seven of the wells in this study (28%) were found to contain coliphage. This suggests that there are less microbially impacted wells than previously thought based upon the new coliphage testing data. The fact that viruses are detected in any drinking water samples is still not comforting. In the most recent CDC report of waterborne disease outbreaks viruses accounted for more illnesses than all of the other pathogens combined (Blackburn et. al. 2004). The number of waterborne outbreaks with an unknown source decreased from 43.6% in the 1999-2000 monitoring period to 22.6% in the 2001-2002 monitoring period (Blackburn et. al. 2004). Improvements in testing for viruses have led to a better ability to identify sources of waterborne illness. Testing for viruses is not part of the current water sampling requirements for potable water. This suggests that there could be many water systems that are not identified as contaminated with the current bacterial indicators that may cause illness in water consumers.

There were significantly fewer new wells with detects of microbes in water samples collected during the year of testing than older wells. This provides some comfort that improvements in well construction methods and increases in minimum casing depth requirements in Door County are improving drinking water quality. Overall, 60% of the wells did not test positive for fecal bacteriological indicators.

Additional site work would help analyze the impact of increasing casing depth and well depth. Information on the geologic features of the dolomite bedrock and prevalence of karst features would allow more accurate site-specific recommendations for well construction. Past studies have attempted to determine some of these features but more would be needed to best compare individual well sites. It is difficult to find a grouping of existing wells of varying well casing depths, in the same area with similar environmental features to accurately assess whether deeper casing improves the water quality. Installing new wells for research is costly but this may be the only way to have new wells of comparable construction methods and varying depths to compare water quality.

New private well owners are unlikely to install shallow wells because of the minimum casing requirements and the increased risk the owner would face for contamination with a shallow well. Private well owners are also unlikely to pay more to install more casing than is required unless there is known contamination problems in existing area wells that comply with the minimum casing requirements. Another problem with sampling private wells is guaranteed access to the site for sampling. Homeowners often volunteer with good intentions but schedule conflicts in their lives or the researchers may make access to

the water system difficult to adhere to a rigid schedule. Again installing wells specifically for research would eliminate the hassle of scheduling conflicts.

However, identifying whether well casing requirements in use in Door County are adequate for protecting groundwater would be useful data to obtain. Minimum casing requirements for specific areas could be adjusted to address more localized concerns based on this data.

It is disturbing that 40% of the wells did test positive for *coliform* and a fecal bacteriological indicator at some time during the year of testing. This suggests that fecal material is occasionally contaminating the aquifer these wells utilize. The highest percentage of wells impacted with fecal bacterial indicators occurred in August at 32% for *enterococci*. The highest percentage of *E.coli* occurred in September at 20% and coliphage was detected at the same level. The GWR implementation plan has not been finalized. A standard has not been defined to judge whether a well is considered contaminated with fecal material for regulatory purposes. This standard will depend on the final GWR and how states implement the rule. A well with one fecal indicator in one sample may not meet the regulatory definition of a fecally contaminated well. However, the fact that 40% of the wells in this study contained fecal material suggests that 40% of the approximately 400 TN wells in Door County are at a greater risk of being classified as contaminated with fecal material.

Some generalized data were used in this study as a compromise to limit costs. The study data support the conclusion that temperature and precipitation influence the contamination of some wells in Door County. The highest percentage of wells impacted with *coliform* (52%) occurred in August which was also the warmest month. *Enterococci* also appear to have a similar increase in response to warmer temperatures.

Precipitation was found to be a significant variable in regression modeling and graphs of some individual well sites suggested that precipitation events were contributing to the transport of contaminants into the groundwater. In future work, it would be beneficial to collect site specific temperature and precipitation data rather than data from one central Door County location. This would remove the averaging of possible variations that may occur across the county from one site to another.

Karst and soil features were not site specific for this study either. This study obtained the number of karst features in a well area from the available layer data for the county. Individual site surveys would provide more accurate information. However, the limited data available did suggest that the five worst water quality well sites had more karst features than the five best water quality wells. This seems logical and provides support for the EPA GWR position that karst areas are hydrogeologically sensitive to fecal contamination.

The GWR will rely on individual water sampling data for each well to evaluate possible upgrades or treatment options that may be needed at specific wells. This study shows site

specific information is important. The water quality varied greatly in the study wells despite the majority of the wells being constructed in the fractured dolostone aquifer.

The study did show some differences in occurrence between *E.coli* and *enterococci*. These differences may require additional study. Some studies have suggested that there is a stronger relationship between *enterococci* in water systems and human illness (Borchardt² et. al. 2003). This suggests *enterococci* may be a better indicator of pathogen contamination than *E.coli*. In the 25 well sites evaluated in this study *enterococci* was detected more often and had a stronger correlation with *coliform*. This suggests that *enterococci* testing would at least be a useful addition to diagnosis of water systems susceptible to fecal contamination that the current *E.coli* testing does not identify.

This study did provide some insight into the continuing water quality concerns in Door County. The GWR will force some changes in how Door County evaluates drinking water quality through testing requirements for public systems. The cost of installing treatment for small TN water systems is still a financial concern for water system owners. Financial assistance is one area that the water system owners could discuss with lawmakers to pursue changes in the funding program that exists. The fecal contamination found in study wells suggests changes are needed in water supplies to provide safe water to the public. The GWR is a step forward in achieving this goal.

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