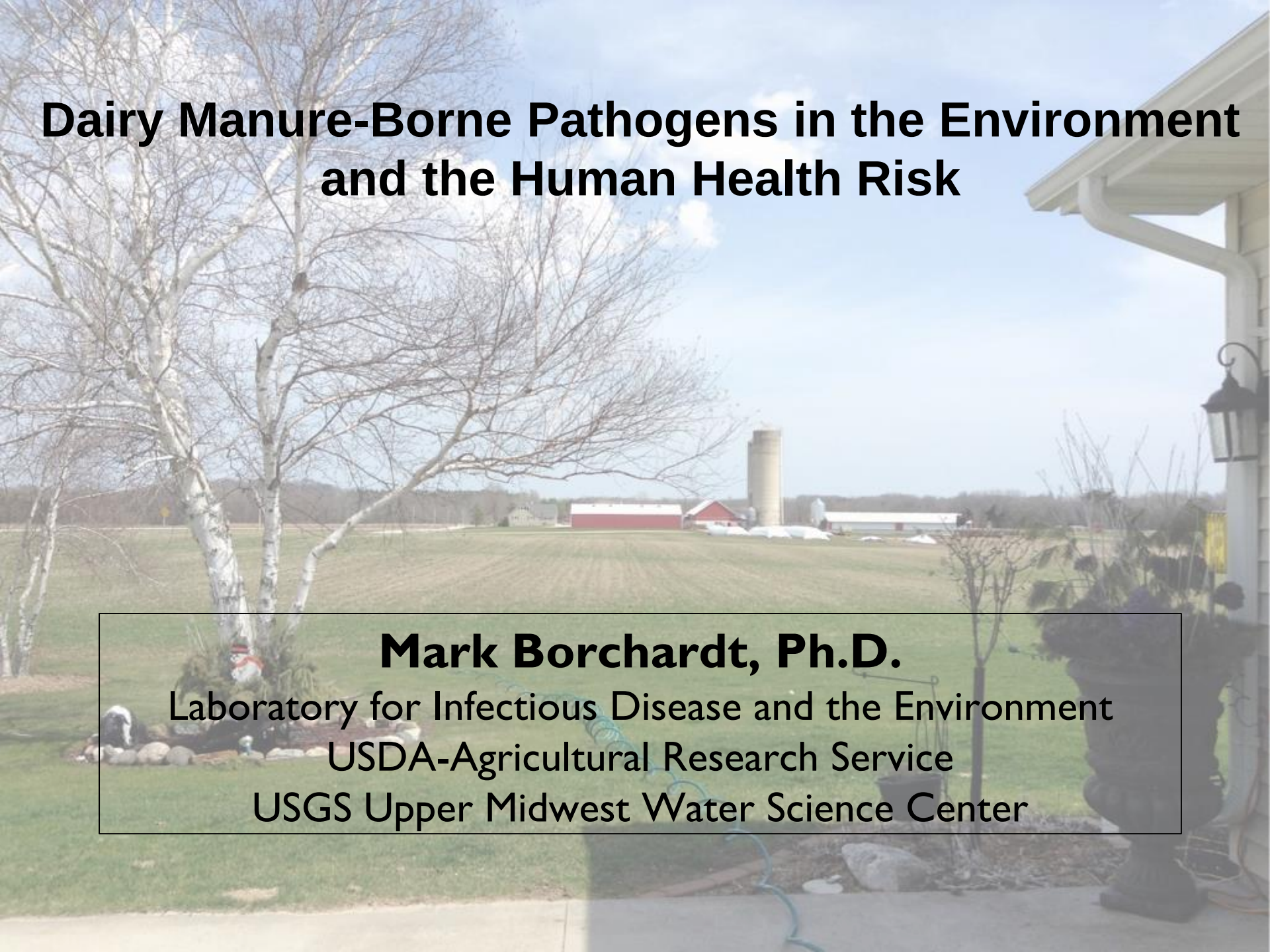
Dairy Manure-Borne Pathogens in the Environment and the Human Health Risk



Mark Borchardt, Ph.D.
Laboratory for Infectious Disease and the Environment
USDA-Agricultural Research Service
USGS Upper Midwest Water Science Center

Manure's Double-Edged Sword

Manure as Asset

Manure field-application is a cost-effective and sustainable approach for optimal soil tilth and fertility

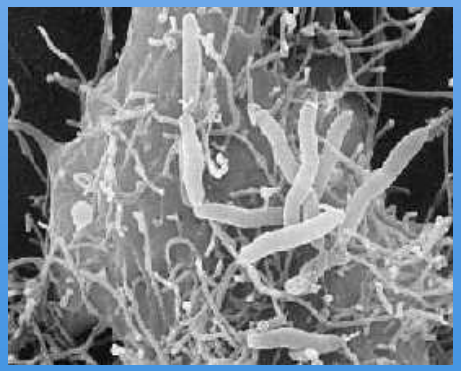
Manure as Liability

Manure may contain pathogens harmful to both humans and livestock

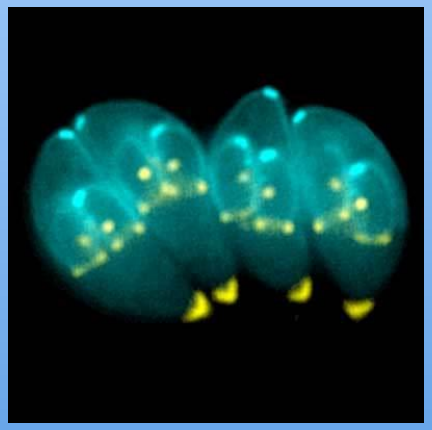
Societal goal: Maximize the beneficial uses of manure while minimizing environmental pathogen transmission

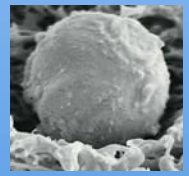
Pathogens Present in Dairy Manure

Bacteria (e.g., *Campylobacter*, *Salmonella*, *E. coli* O157:H7, *Listeria monocytogenes)*

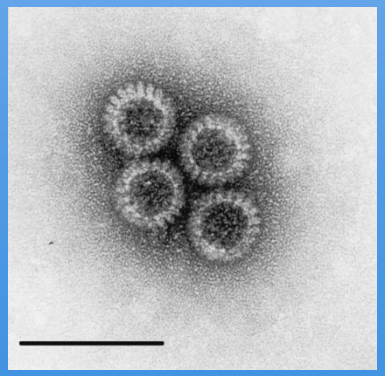


Protozoa, (e.g., *Cryptosporidium*, *Giardia*, *Toxoplasma)*

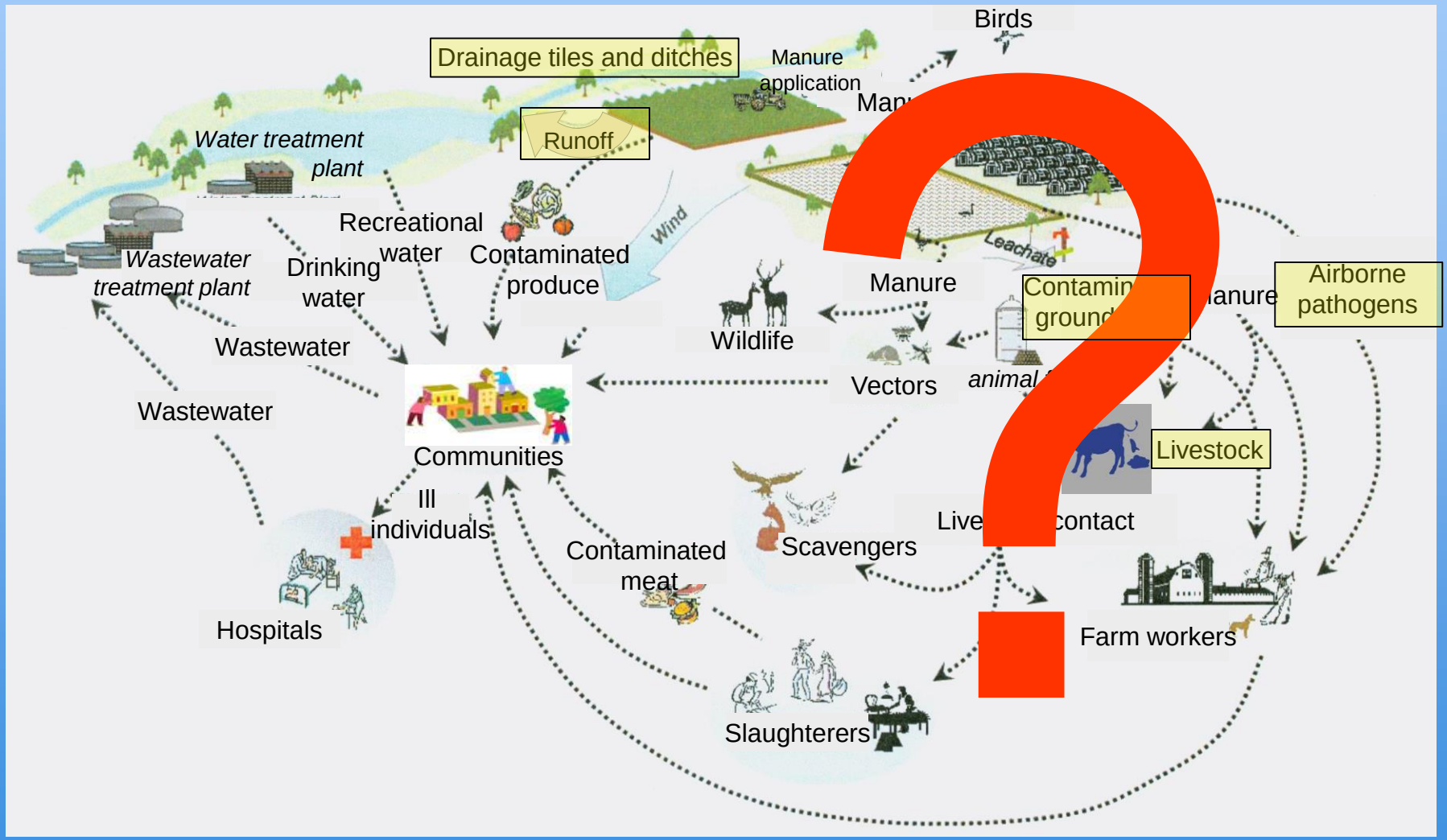




Viruses (e.g., rotavirus)



Human and Livestock Pathogen Movement in the Environment



Rogers and Haines, 2005, EPA/600/R-06/021

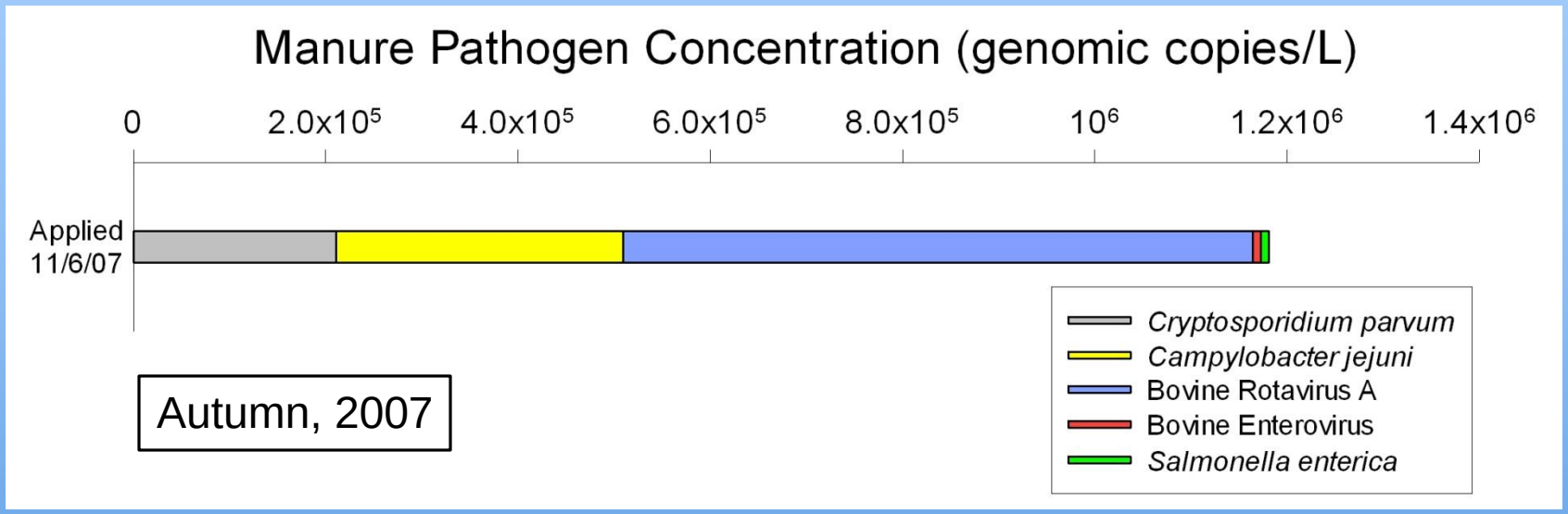
Estimates of Enteric Illness Attributable to Contact with Animals and Their Environments in the United States

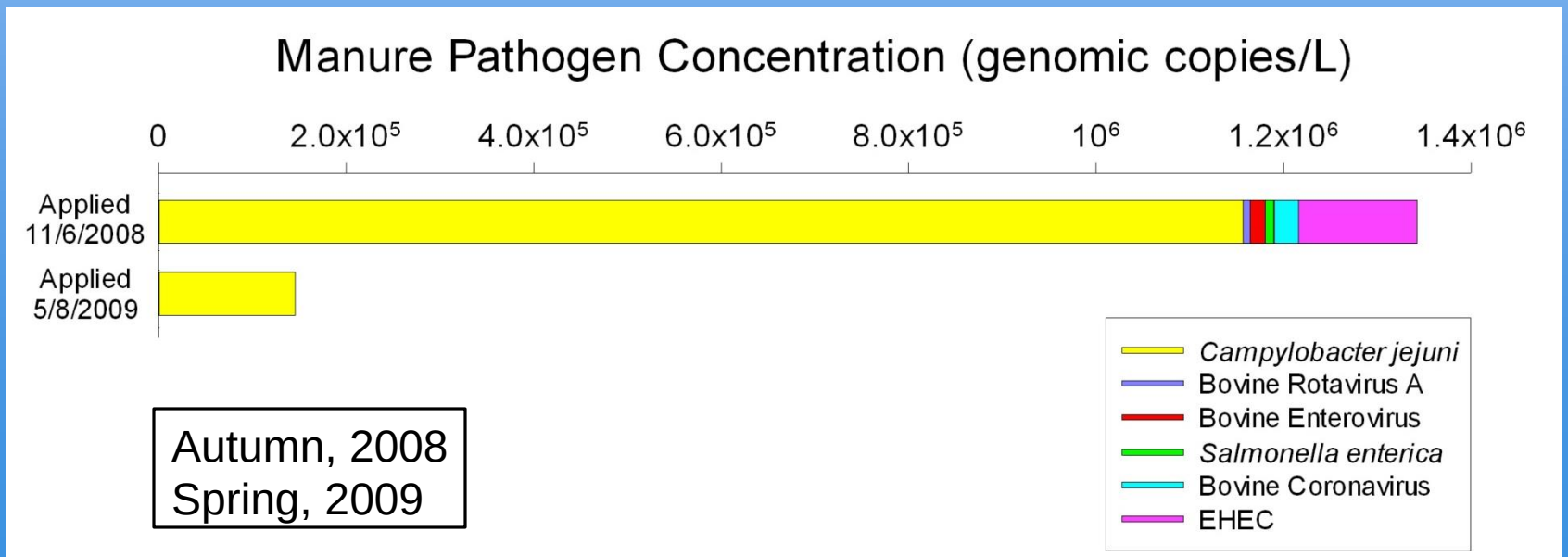
- 445,213 /3.2 million illnesses (14%) from animal contact

Organism	% from animal contact	Annual # illnesses	Annual # hospitalizations	Annual # deaths
Campylobacter species	17%	187,481	1,877	17
ST E. coli	14	16,057	230	2
Non-typhoid Salmonella	11	127,155	2,392	47
Cryptosporidium species	16	113,344	412	7

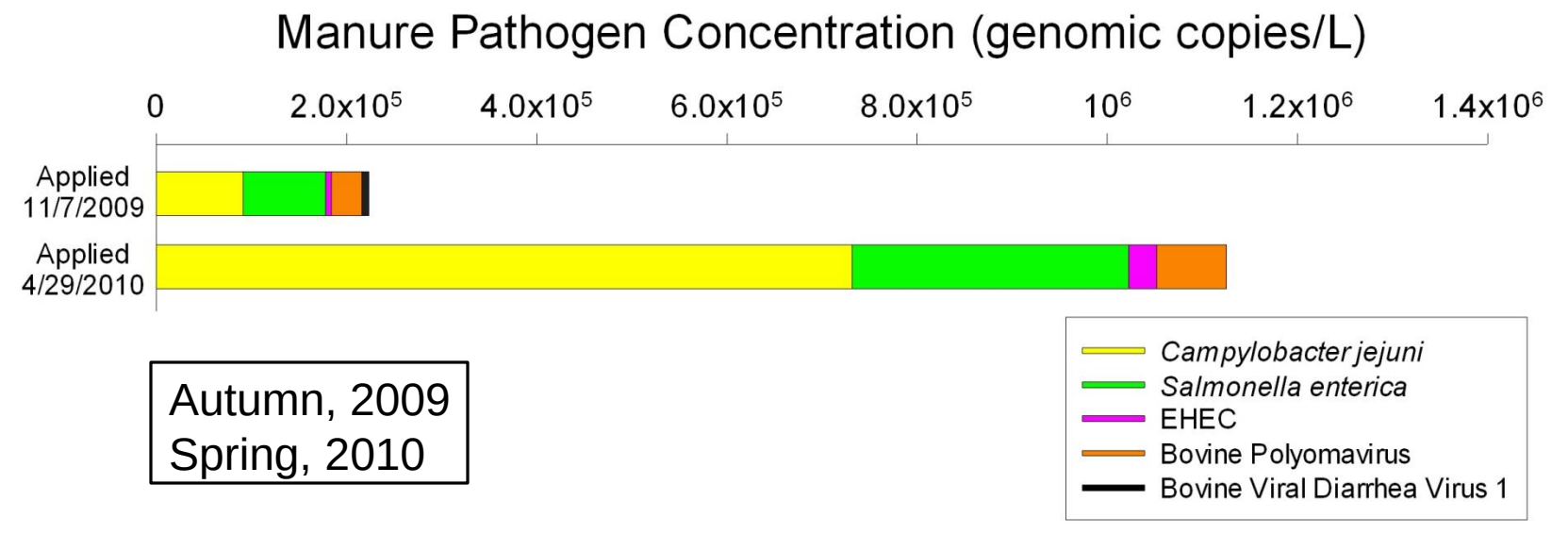
Centers for Disease Control and Prevention: Hale et al. 2012, CID, 54:S472-79.

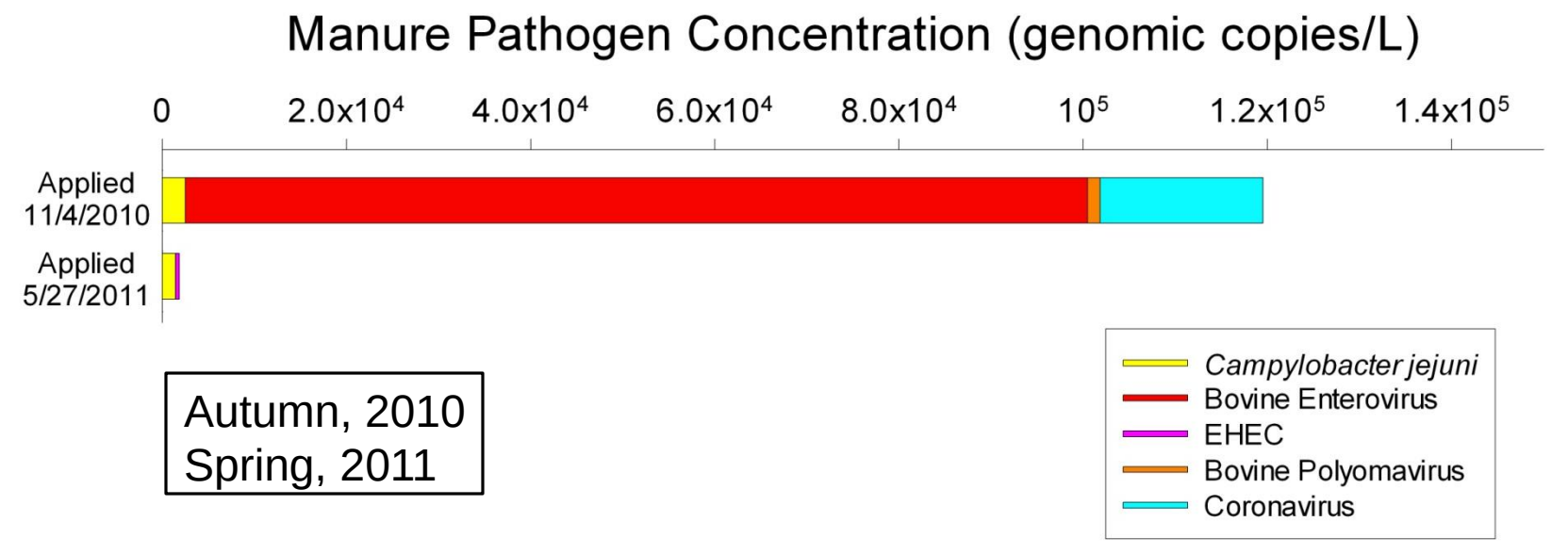
Pathogens in manure from a single farm by year and season





Pathogens in manure from a single farm by year and season





Brown Water Events in Northeast Wisconsin

- Groundwater recharge, especially spring snow melt, can generate brown water events
- Several outbreaks associated with these events e.g., EHEC, *Campylobacter jejuni*
- This well is code compliant, 123 ft deep, cased to 63 ft

Photos courtesy of Chuck Wagner

Kewaunee County Cattle

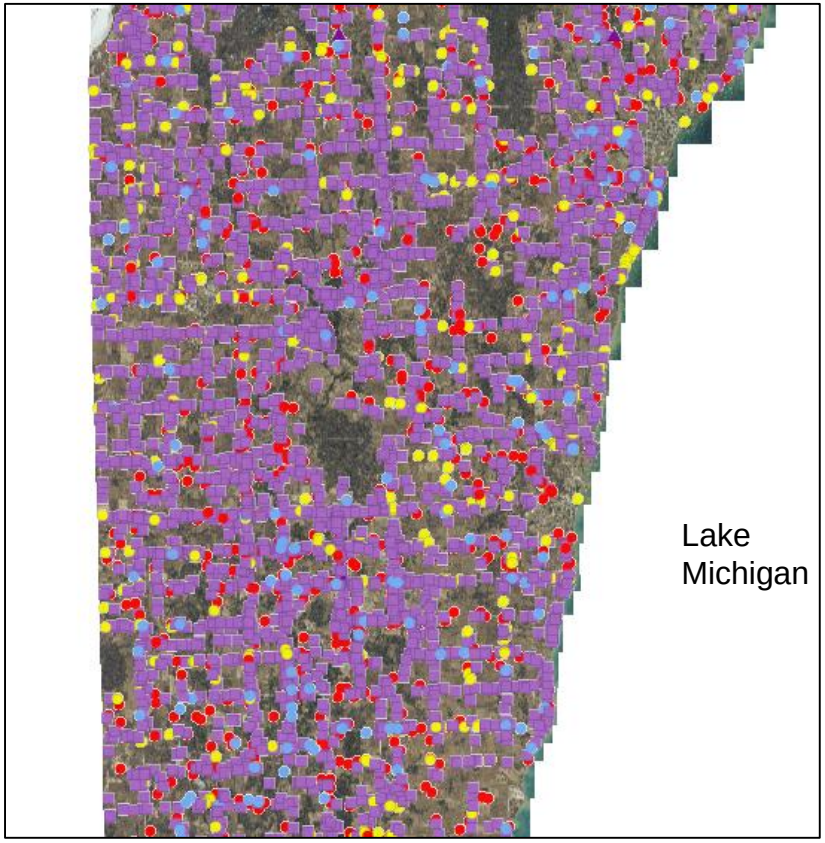
- All cattle & calves in 2016 = 97,000
- Milk cows in 2013 = 45,500
- Milk cow herds in 2016 = 167
- Concentrated Animal Feeding Operations (CAFOs) 15 dairy, one beef
- Approximately 700 million gallons cattle manure per year

Kewaunee County Septic Systems

- 4822 septic systems in the county
- 540 holding tanks, 155 abandoned

Personal comm. Lee Luft, Kewaunee County Supervisor, March 7, 2017

Legend
Purple = replaced or inspected
Red = not inspected
Yellow = holding tank
Blue = abandoned system



Kewaunee County septic systems

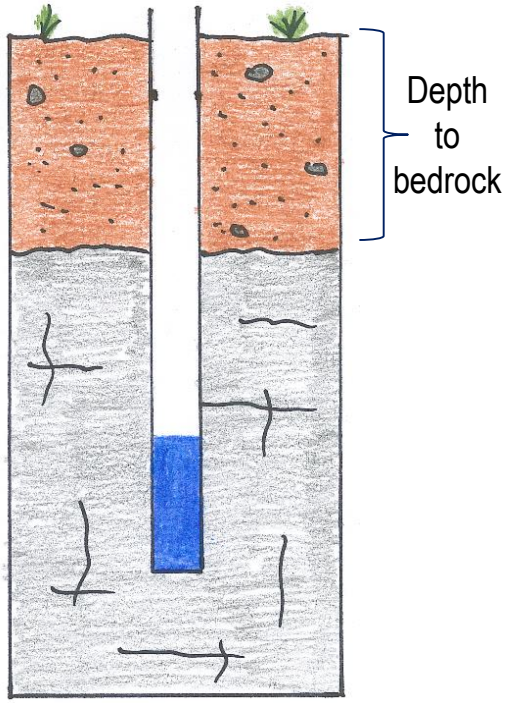
Approximately 200 million gallons septic effluent per year released to the subsurface

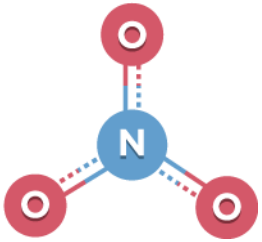
Research Objectives

1. Estimate county-wide contamination rate for nitrate and indicator bacteria as related to depth-to-bedrock
2. Determine source of fecal contamination using viruses and fecal markers
3. Identify risk factors for private well contamination using statistical models

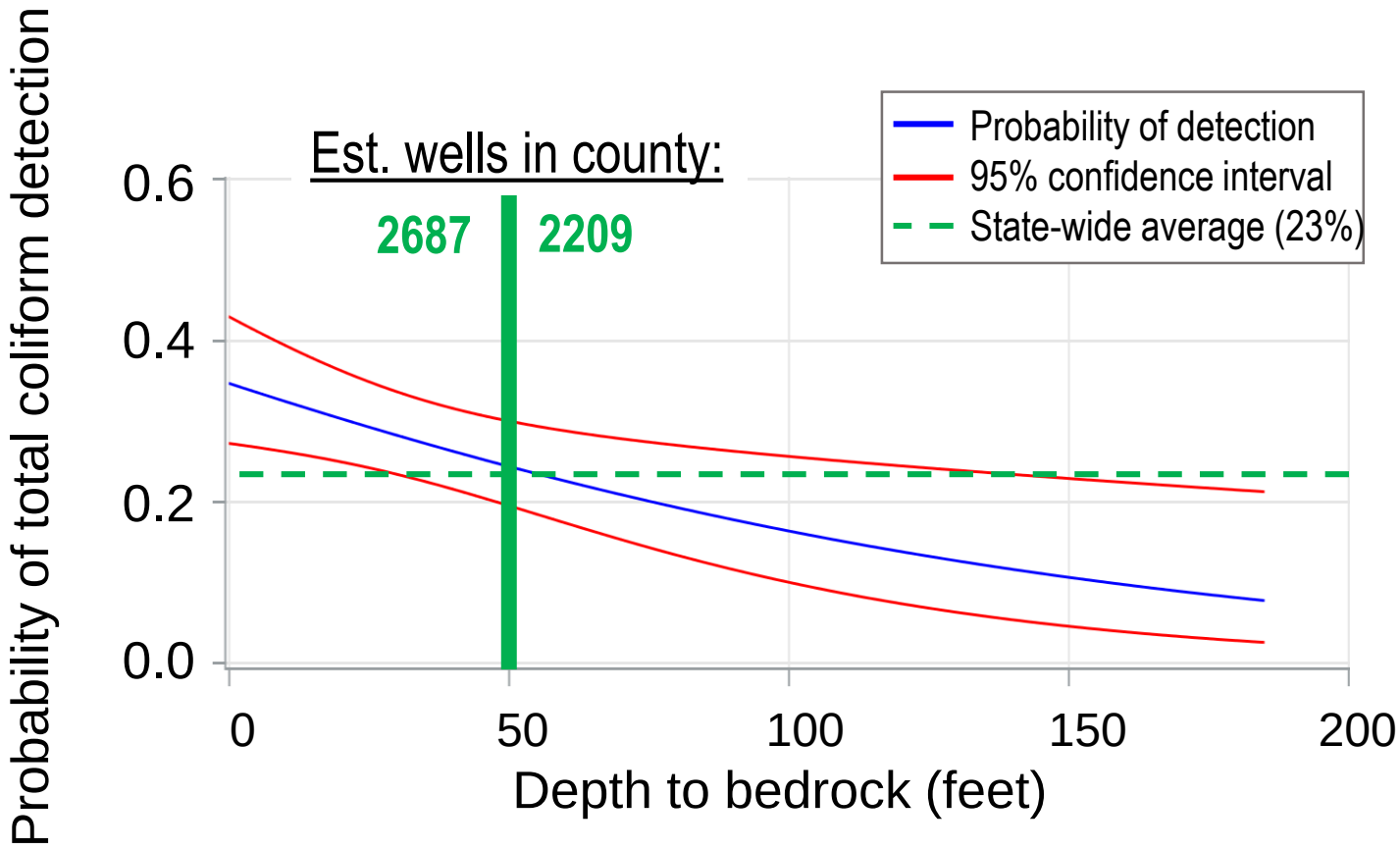
Objective 1: Total Coliform, *E. coli*, Nitrate

- County-wide randomized sampling of private wells – 4,896 in county
- Stratified by depth-to-bedrock: 0-5 ft, 5-20 ft, 20+ ft
- Participation rate ~ 50%
- Several day "Synoptic" sampling
- Recharge
  - November 2015
  - 317 wells in analysis
- No recharge
  - July 2016
  - 400 wells in analysis





Effect of depth to bedrock on total coliform contamination



Fall, 2015

n = 315

p = 0.009

Objective 2: Determine fecal source

- Randomized stratified sampling from 208 wells positive for total coliform, *E. coli*, or high nitrate ($N\text{-}NO_3^-$ > 10 ppm)
- Five sampling rounds:
    - April, August, November, 2016
    - January and March, 2017

Study Sampling and Analyses

- Collected 138 samples from 131 household wells in Kewaunee County
- Pump ~800 L through hemodialysis filters
- qPCR for microbial genetic targets
  - Human-specific microbes
  - Bovine-specific microbes
  - Non-specific microbes (pathogens of both people and cattle)

Microbes: Identifying the Fecal Source

(n = 138 samples from 131 wells) (red font indicates pathogenic)

Host	Microorganism	Wells	Concentration (gene copies/L)
Human-specific	Adenovirus A	1	1
	Bacteroidales-like Hum M2	7	< 1 – 1050
	Human *Bacteroides*	27	< 1 – 34
	Cryptosporidium hominis	1	qualitative
	Rotavirus A (G1 P[8])	7	qualitative
	All	**33**	

Not detected: [human-specific] adenovirus B & C, D, F, enterovirus, human polyomavirus, norovirus GI & GII
[bovine-specific] coronavirus, bovine diarrheal virus 1 & 2

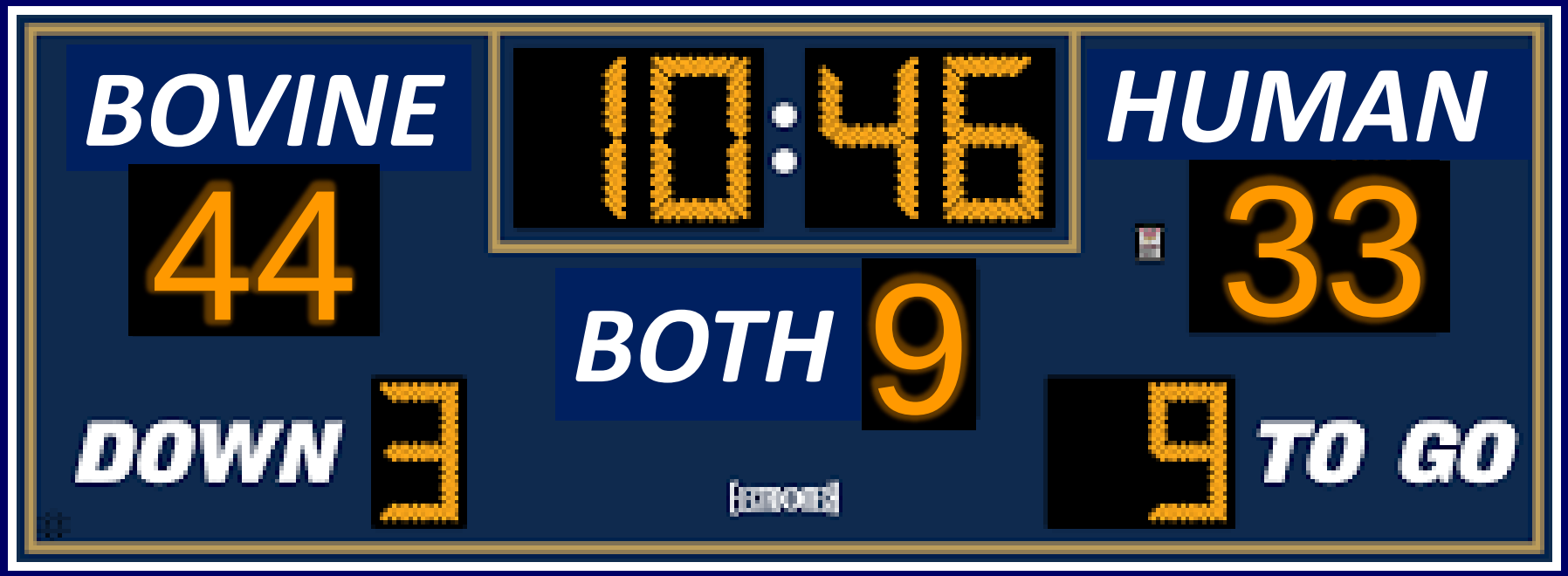
Microbes: Identifying the Fecal Source

(n = 138 samples from 131 wells) (red font indicates pathogenic)

Host	Microorganism	Wells	Concentration (gene copies/L)
Bovine-specific	*Bacteroidales*-like Cow M2	2	29 - 915
	Bacteroidales-like Cow M3	4	3 – 49,818
	Bovine *Bacteroides*	36	< 1 – 42,398
	Bovine polyomavirus	8	< 1 – 451
	Bovine enterovirus	1	2
	Rotavirus A (G10 P[11])	12	qualitative
	All	**44**	

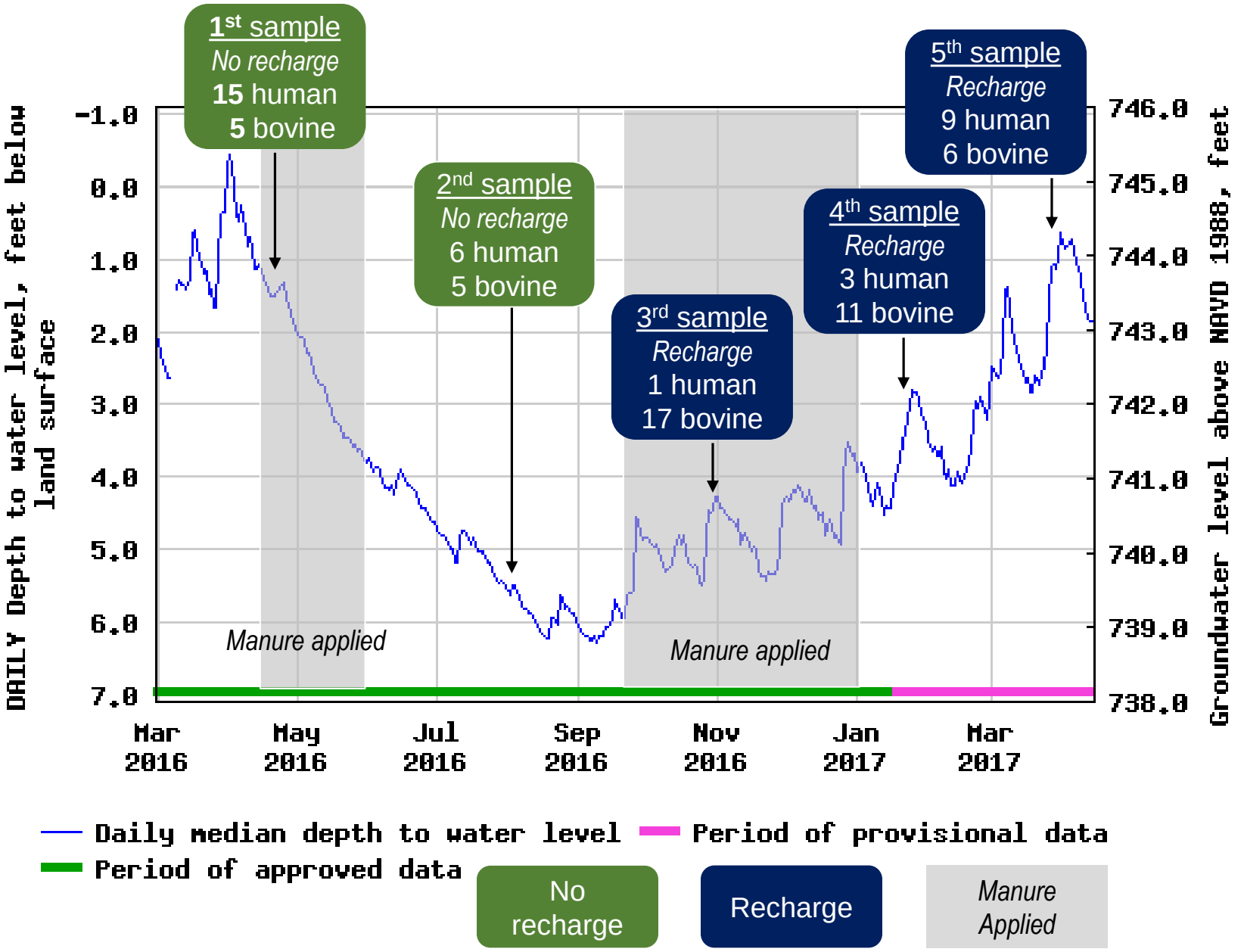
Not detected: [human-specific] adenovirus B & C, D, F, enterovirus, human polyomavirus, norovirus GI & GII
[bovine-specific] coronavirus, bovine diarrheal virus 1 & 2

Well Contamination Scoreboard



Host	Microorganism	Wells	Concentration (gene copies/L)
Non-specific	*Campylobacter jejuni*	1	< 1
	Cryptosporidium parvum	13	qualitative
	Cryptosporidium spp.	16	< 1 – 3
	Giardia lamblia	2	< 1
	Pathogenic *E. coli* (*eae* gene)	1	4
	Pathogenic *E. coli* (*stx1* gene)	1	16
	Pathogenic *E. coli* (*stx2* gene)	1	1
	Pepper mild mottle virus	13	2 - 3811
	Rotavirus A (*NSP3* gene)	17	< 1 – 4481
	Rotavirus A (*VP7* gene)	7	< 1 – 732
	Rotavirus C	3	45 – 1301
	Salmonella (*invA* gene)	3	< 1 – 13
	Salmonella (*ttr* gene)	5	5 – 59
	All	**44**	
	Total positive wells	**79**	**< 1 - 49818**

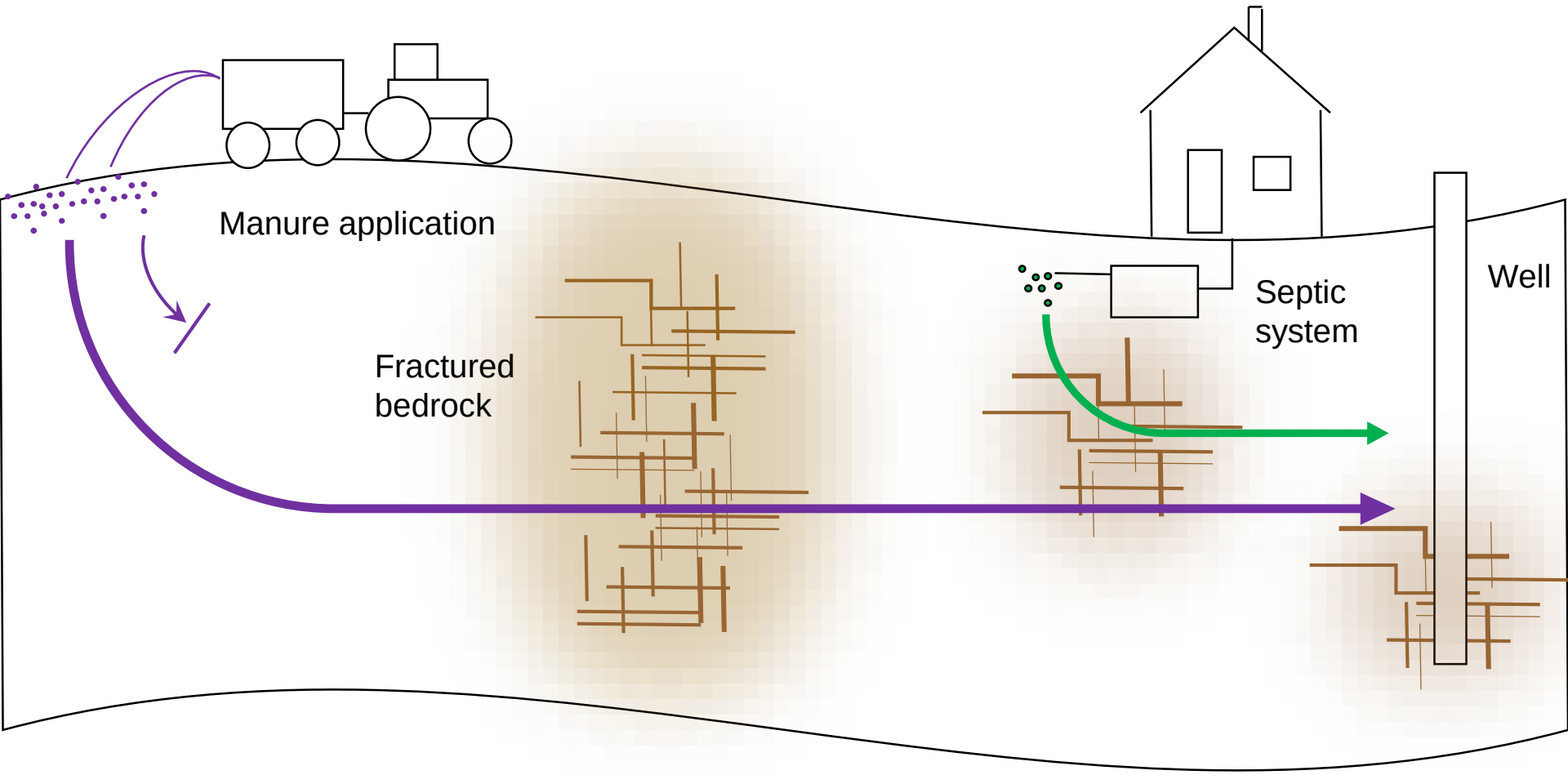
Groundwater levels during sampling for pathogens & fecal indicators



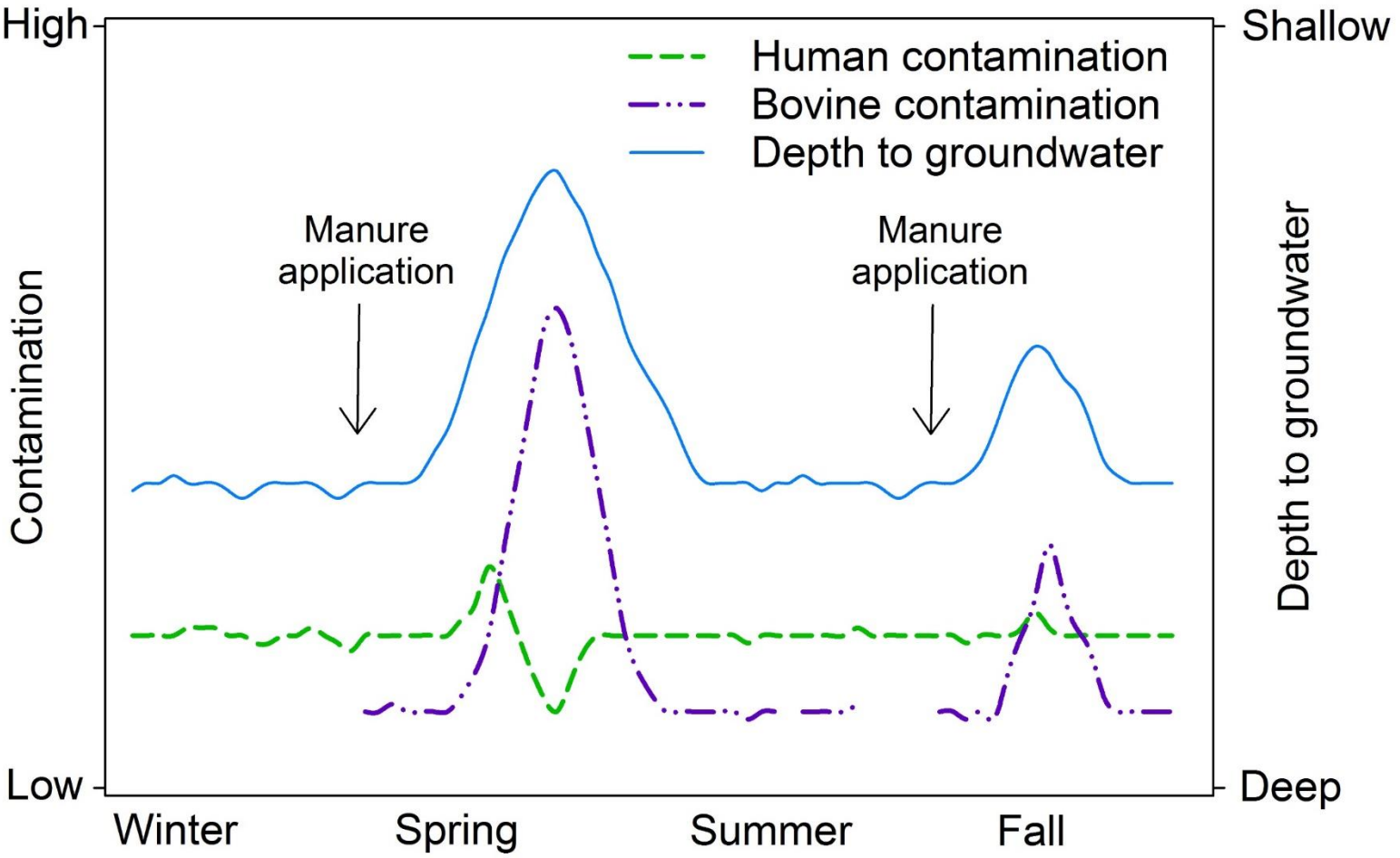
Conceptual Model of Fecal Contamination in Kewaunee County - 1

Bovine pathogen source
- Large fecal source
- Surface applied periodically
- Episodic infiltration

Human pathogen source
- Small fecal source
- Sub-surface release continuously
- Continuous infiltration



Conceptual Model of Fecal Contamination in Kewaunee County - 2



Objective 3: Statistical Risk Factor Analysis

Contamination Sources – *Distance; count or acres within 750, 1500, 3000 ft of well*

Bovine	*Human*
Agricultural fields	Septic systems (all, drain field, uninspected)
Manure storages	Septage-applied fields

Karst Features – *Count within 750, 1500 and 3000 feet of well*

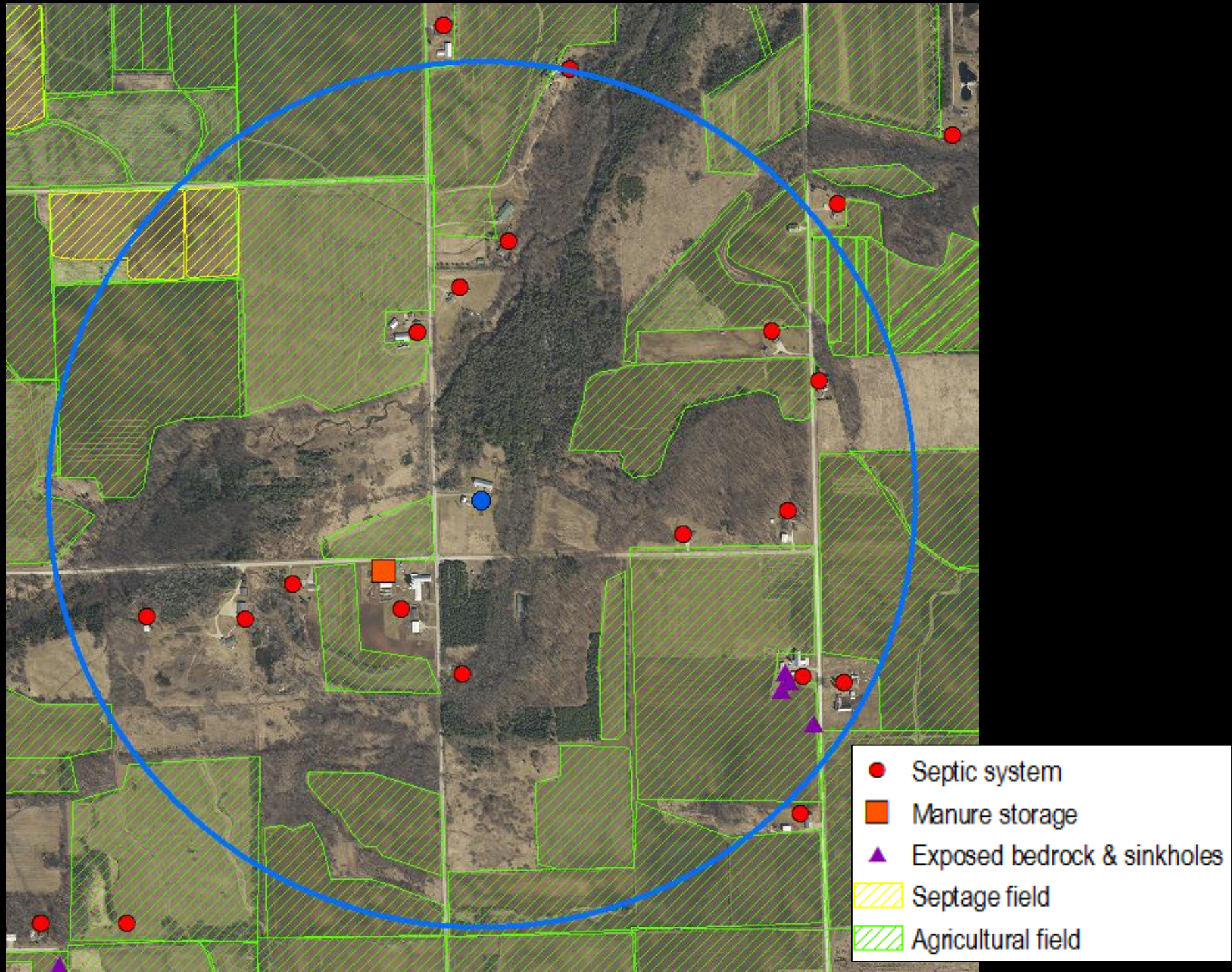
- Sinkholes
- Bedrock features at the surface

Precipitation & Groundwater Recharge – *2, 7, 14, 21 days prior to sampling*

- Precipitation (cumulative, no snowfall)
- Groundwater recharge (cumulative)
- Depth to groundwater (median & minimum)

Well Construction & Siting

- Well depth
- Casing depth
- Depth to groundwater
- Depth to bedrock
- Open interval length
- Well age
- Length of casing into bedrock
- Length of casing below water table
- Elevation
- Soil drainage


Septic system
Manure storage
Exposed bedrock & sinkholes
Septage field
Agricultural field

Risk Factors for Human Fecal Contamination

Important factors

- Distance to nearest neighbor's septic system
- Number of neighbors' septic drain fields around well
- Precipitation
- Depth to groundwater
- Depth to bedrock

Surprising unimportant factors

- Well construction variables (e.g., casing depth)

Risk Factors for Manure Contamination

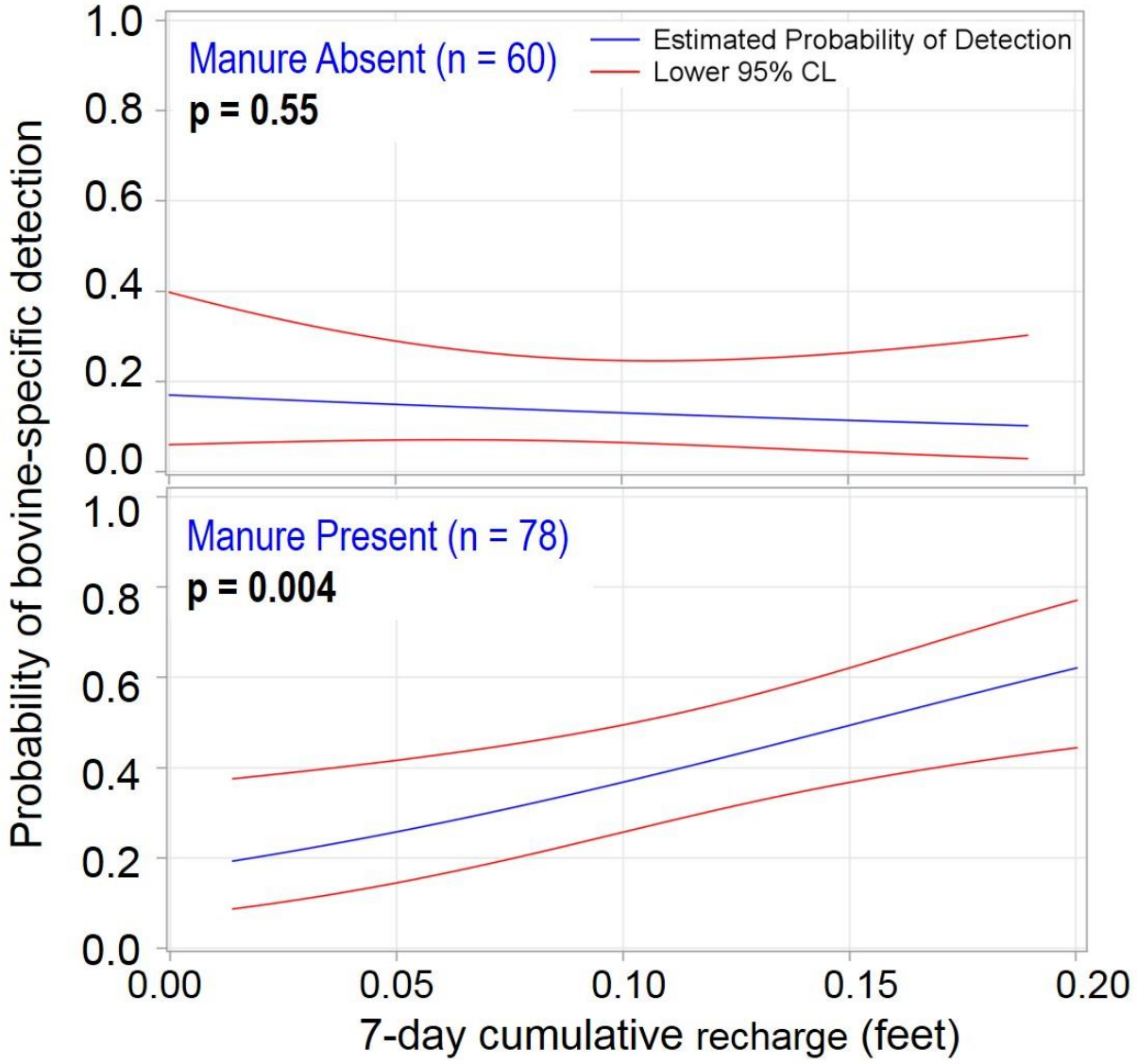
Important factors

- Recharge
- Depth to groundwater
- Depth to bedrock
- Interactions between ag land use, manure application, and groundwater recharge

Surprising unimportant factors

- Well construction variables (e.g., casing depth)

Bovine-Specific Microbes in Private Wells by Previous 7 Days Cumulative Recharge



Risk Factors for Total Coliform and High Nitrate Contamination

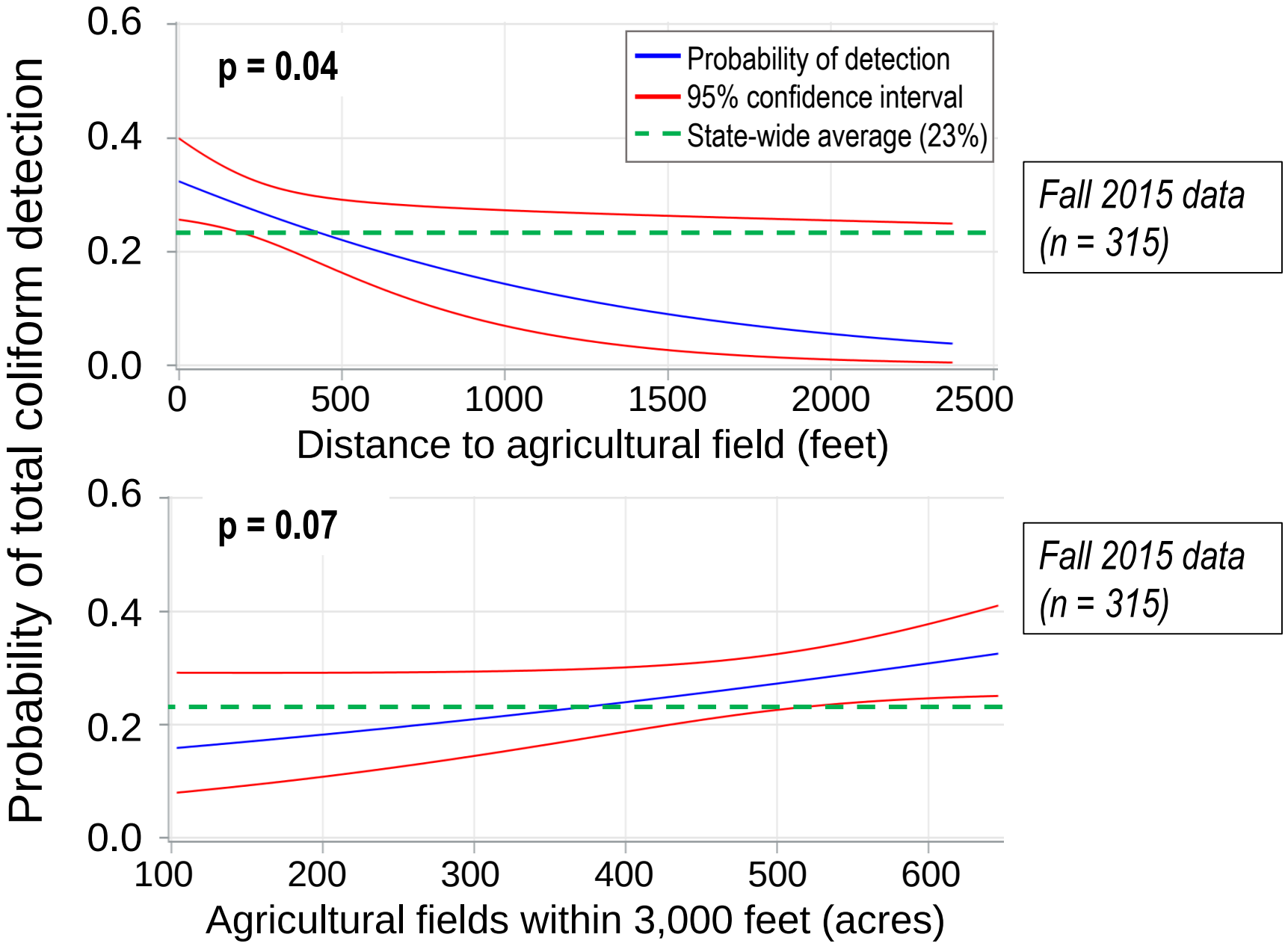
Important factors

- Distance between well and agricultural field
- Number of acres of ag fields around well
- Number of sink holes around well
- Distance to nearest manure lagoon
- Recharge
- Depth to bedrock

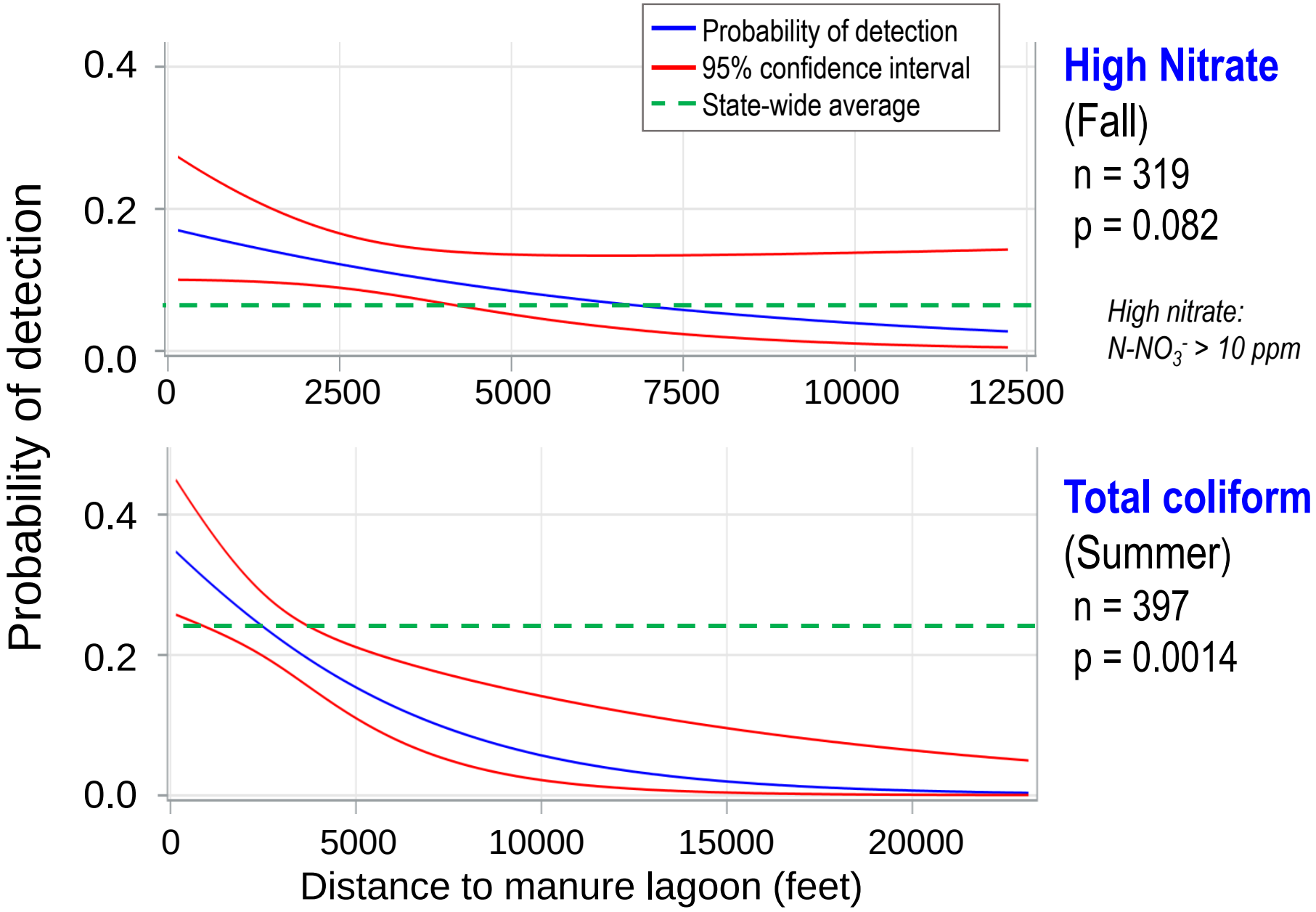
Surprising unimportant factors

- Septic system variables
- Well construction variables (e.g., casing depth)

Risk Factors for Total Coliform Contamination



Manure Lagoon Association with Coliforms and Nitrate



Do people get sick from drinking contaminated private well water?

- Consider one pathogen: *Cryptospordium parvum*
- Confirmed cryptosporidiosis cases in Kewaunee County reported to State:
  - 2 to 9 cases per year (2010 to 2016)
- Under-reporting of cryptosporidiosis cases in the USA is estimated to be 100-fold (Centers for Disease Control and Prevention, 2012)
- Therefore, in Kewaunee County there are likely 200 to 900 cryptosporidiosis cases per year

How many of these cases are from private wells?

Estimate of Kewaunee County *Cryptosporidium parvum* infections from private wells

	People	Calves
Population using private wells	12,200	17,300
Wells contaminated by *C. parvum*	3.1%	3.1% (assumed)
Population exposed per day	380	540
Infections per exposure	10 infections per 10,000 people	85 infections per 10,000 calves
Total infections per year	140	1,700

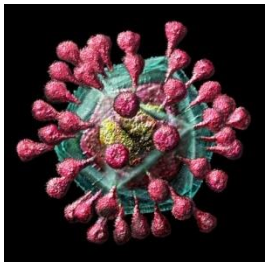
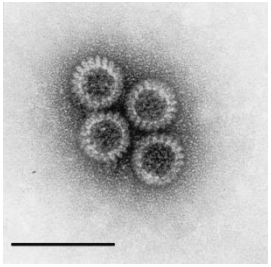
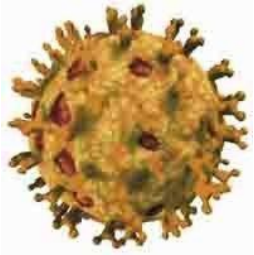
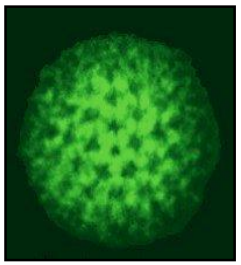
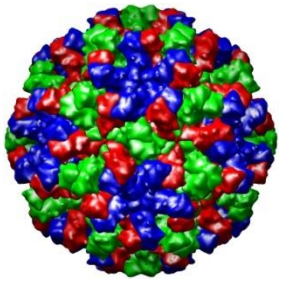
Solutions for Preventing Exposure to Manure-borne Pathogens in the Environment

Practices to Minimize Transport	Practices to Maximize Inactivation
Distance between livestock and waterways	Storage time
Vegetated treatment areas	Chemical treatment (e.g., lime)
Settling basins and wetlands	Thermophilic processes (e.g., aerated composting)
Manure storage and treatment lagoons	Anaerobic digestion

From: Atwill et al. 2011 An Introduction to Waterborne Pathogens in Agricultural Watersheds, NRCS

Questions?

Comments?

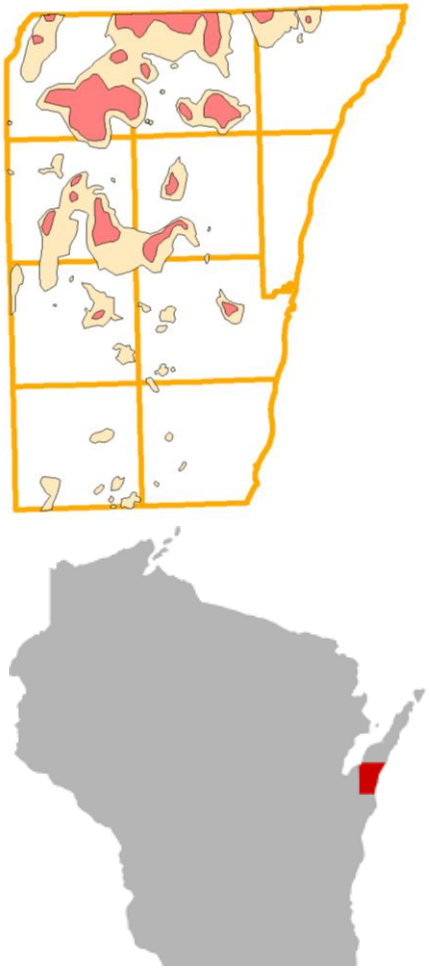






County-wide contamination rate; weighted by depth to bedrock

Percent wells contaminated

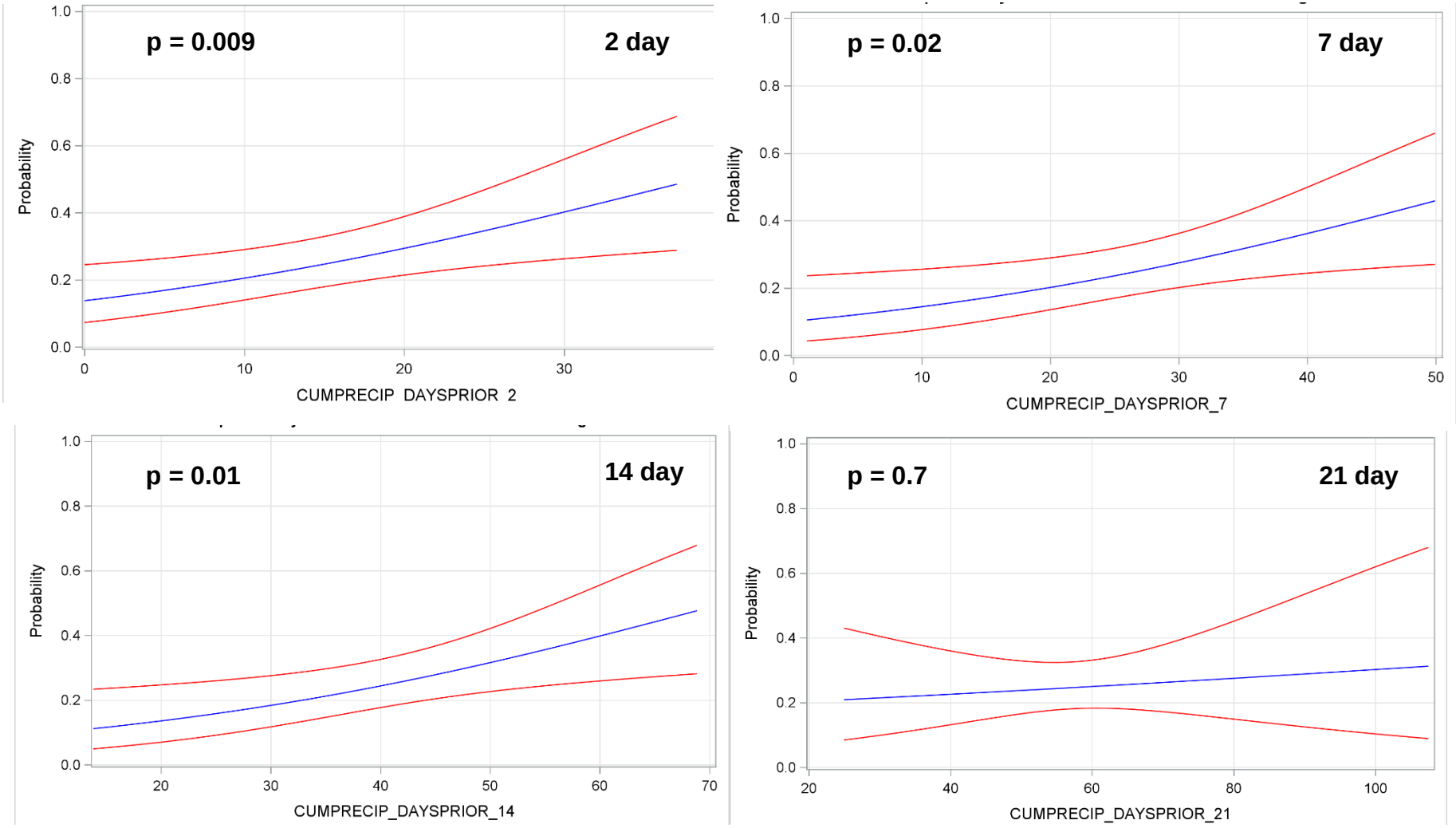
	Kewaunee County		Wisconsin*
	Recharge (n = 317)	No Recharge (n = 400)	(n = 534)
Total coliform	20.8	22.2	22.8
E. coli	0.4	1.2	2.6
High nitrate	7.4	6.8	6.6
Any of the 3 contaminants	26.4	27.6	NA

High nitrate: exceeds health standard; N-NO_3^- > 10 ppm



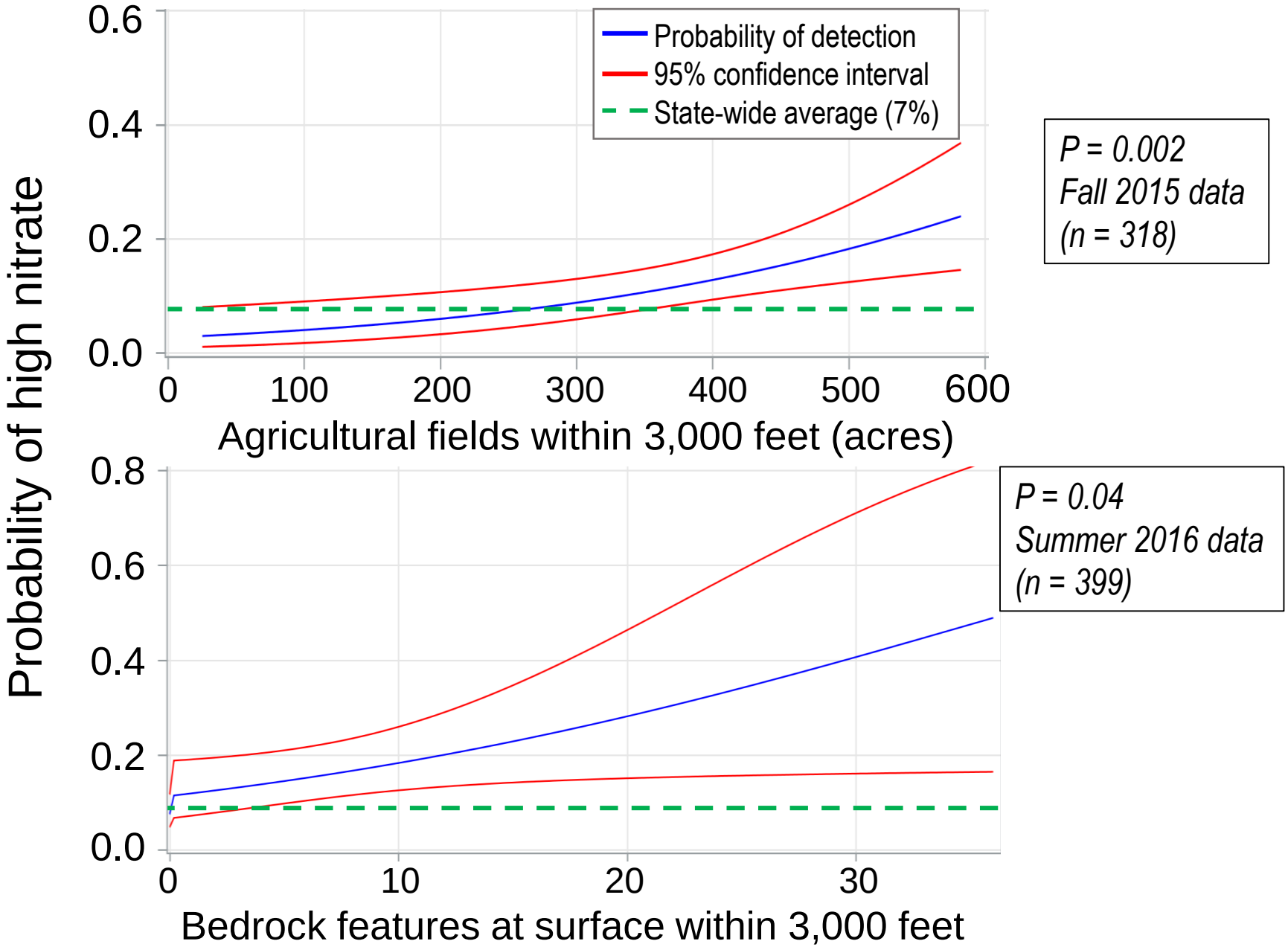
*private wells sampled; *Information on the quality of water found at community water systems and private wells. United States GAO/RCED-97-123, June 1997*

Probability of well contamination with human fecal markers as related to cumulative precipitation



Cumulative precipitation in millimeters

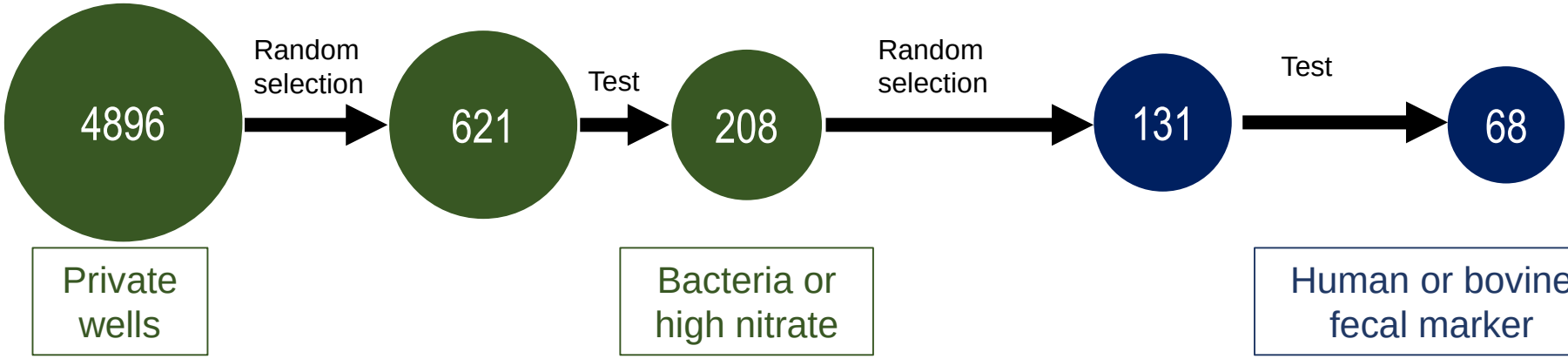
Risk Factors for High Nitrate ($N\text{-}NO_3^- > 10$ *ppm)* Contamination



Project objectives & study design

1. Measure total coliform, *E. coli*, nitrate → 2. Determine fecal source

Given contamination



Outcome: County-wide occurrence as % wells contaminated

Outcome: Number of wells with human or bovine fecal markers

Pathogens & fecal markers in Kewaunee County: Comparison to other studies

Wisconsin 2003:
Private wells
50 tested

Ontario 2017:
Private wells
11 tested

Pennsylvania 2017
Private wells
5 tested

0% 8% 15% 45% 60% 100%

Study wells positive for microorganisms

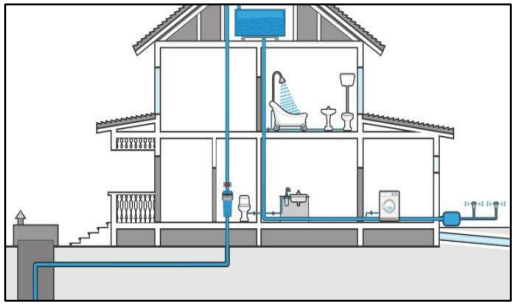
Canada & USA 1990 – 2013
Public & private wells
12,616 tested

Kewaunee County
Private wells
131 tested

From Farm Field to Household Well

Manure applied Oct 25, 2016

> 1 inch rain Oct 26, 2016



House near field

Farm field sampled Oct 27, 2016

Bovine Bacteroides
Bovine enterovirus
Bovine polyomavirus
M2 Bacteroides-like
M3 Bacteroides-like

Rotavirus A NSP3
Rotavirus A VP7
Rotavirus C

Tap water Oct 27, 2016

Bovine Bacteroides
Bovine enterovirus
Bovine polyomavirus
M2 Bacteroides-like
M3 Bacteroides-like
Campylobacter jejuni
Cryptosporidium
Rotavirus A NSP3
Rotavirus A VP7
Rotavirus C

From Farm Field to Household Well

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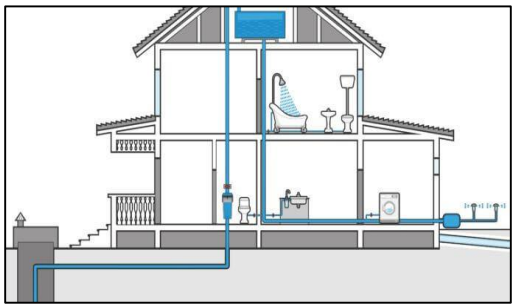
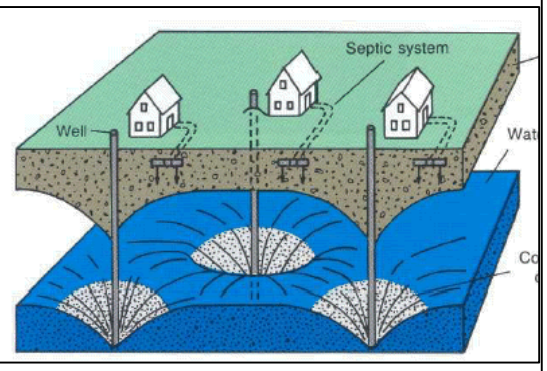


House near field

Neighbor's well sampled Oct 31, 2016



Bovine Bacteroides

Bovine polyomavirus
M2 Bacteroides-like
M3 Bacteroides-like

Rotavirus A NSP3
Rotavirus A VP7
Rotavirus C

Tap water Oct 27, 2016

Bovine Bacteroides
Bovine enterovirus
Bovine polyomavirus
M2 Bacteroides-like
M3 Bacteroides-like
Campylobacter jejuni
Cryptosporidium
Rotavirus A NSP3
Rotavirus A VP7
Rotavirus C

Summary

- Well contamination in the fractured dolomite aquifer in Kewaunee County results from both human and bovine fecal sources.
- Wells are contaminated with pathogens of significant concern: *Salmonella*, EHEC, *Cryptosporidium*, rotavirus.
- At depths to bedrock less than 50 feet total coliform and nitrate contamination rates generally exceed statewide averages.
- Risk factors for well contamination are: groundwater recharge, depth to groundwater, sink holes, precipitation, timing of manure application, agricultural land use, and the density of septic drain fields.

We Thank…

Kevin Masarik
UW-Stevens Point Watershed Science and Education

Kevin Erb
UW-Extension – Environmental Resources Center

Leah Kammel & Laura Hubbard
Upper Midwest Water Science Center

Stephen Mauel
Wisconsin Geological & Natural History Survey

Dustin Goering and Liz Houle
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