Assessing Levels and Potential Health Effects of Endocrine Disrupting Chemicals in Groundwater Associated with Karst Areas in Northeast Wisconsin

Angela Bauer, Ph.D.
Professor, Human Biology
and Environmental Science and Policy
-and-
Michael Zorn, Ph.D.
Associate Professor, Natural and Applied Sciences, Chemistry, and Environmental Science and Policy

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History of groundwater contamination in northeast Wisconsin

- Areas characterized by carbonate bedrock, shallow soil depths and karst features
- NEW Karst Task Force (2007) reports a significant proportion of wells contaminated by bacteria or high levels of nitrate (~30% in Brown and Calumet Counties)
- Linked to manure runoff, especially during spring thaw

Objectives of study:

- Assess whether the following contaminants are present in well water samples collected in NEW:
  - Bacteria (*E. coli*, total coliform, enterococci)
  - Nitrates
  - Endocrine disrupting contaminants (EDCs)
- Determine whether the presence of EDCs correlates with that of other contaminants
- Determine whether the presence of contaminants in well water correlates with season / recharge events

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What are Endocrine Disrupting Chemicals (EDCs)?

- chemicals that interfere with the normal functioning of hormones in animals and humans
  - Source: animal waste, human waste, pesticides
- potential mechanisms of action:
  - hormone mimicry
  - disturbance of hormone production and secretion
  - disruption of hormone receptors

Dutch, UWGB

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Possible EDC effects in humans

- overall lifetime exposure to estrogens correlates w/ increased risk of cancer in hormone-sensitive tissues
  - prostate cancer, testicular cancer, breast cancer
- developmental problems
  - abnormal development of male sex organs
  - reduced sperm count

How do EDCs get into the water?

- Groundwater: infiltration of run-off into soils
  - stormwater run-off
  - agricultural run-off
    - Synthetic or endogenous hormones
    - Pesticides
  - Leaky septic tanks, landfills

Cattle given estradiol as a growth hormone:
Urine 5-6 x normal concentration of estradiol!

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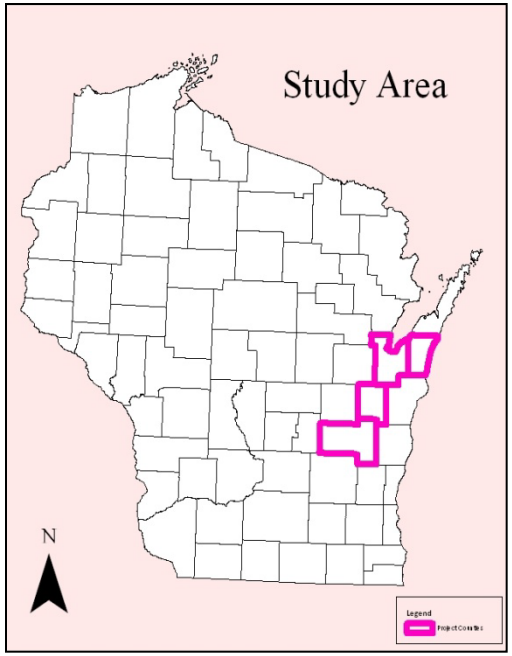
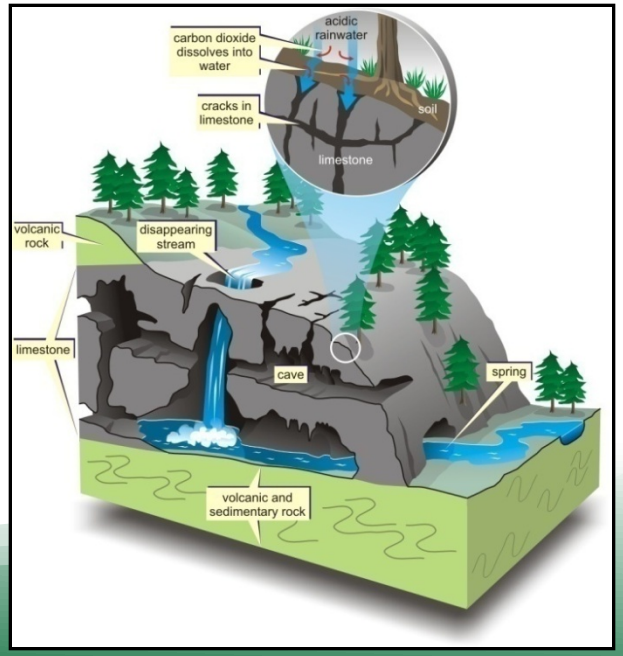
Study Area



- northeast Wisconsin
  - Brown, Calumet, Fond du Lac, & Kewaunee
  - carbonate bedrock, shallow soil depths
    - limestone dissolution → karst features
    - sinkholes, bedrock fractures
  - heavy agriculture
  - wells w/ past contamination
    - bacteria and nitrate
    - pharmaceuticals, hormones?



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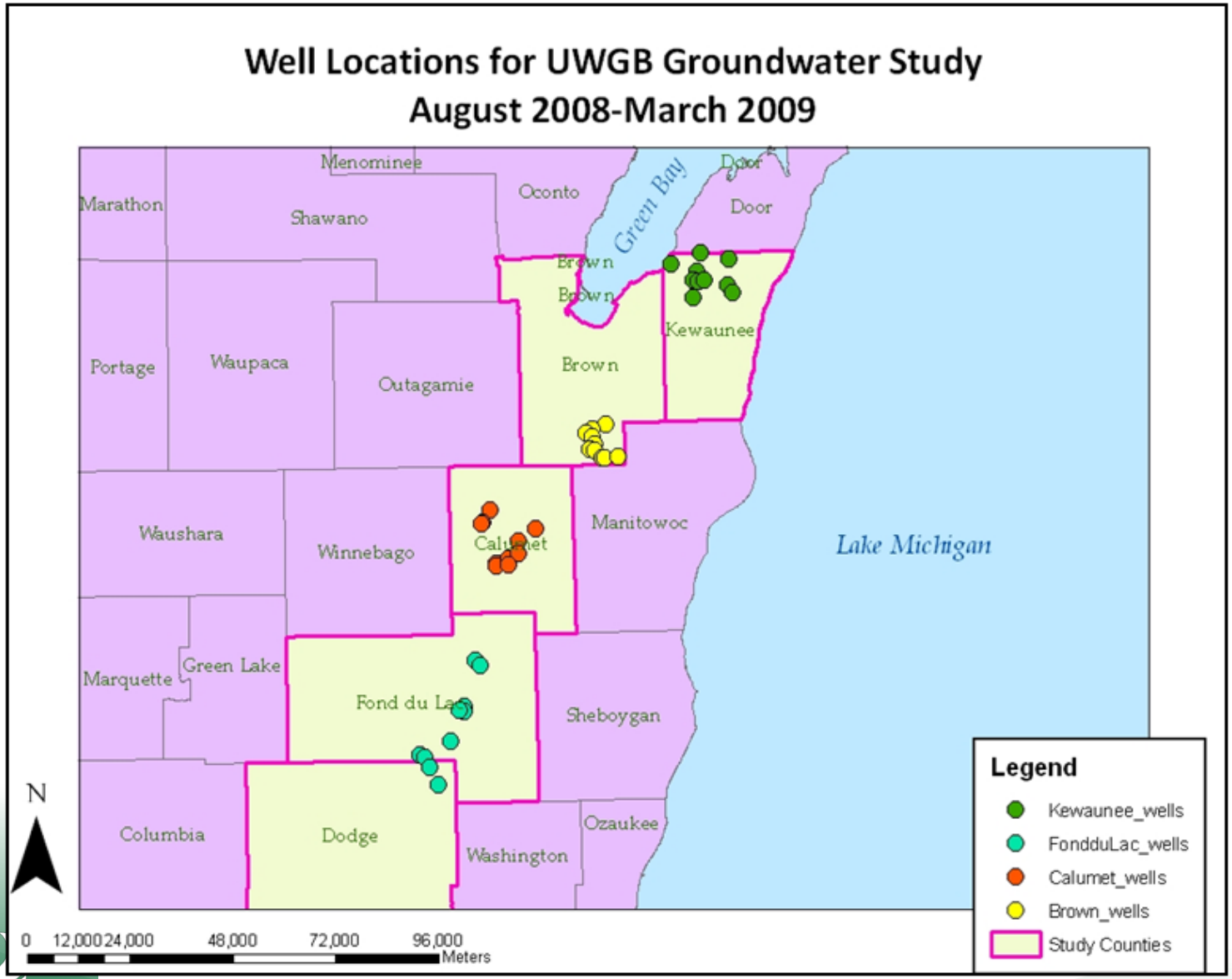

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Methods

- 10 private wells from each county
  - 2 control
  - 8 susceptible
    - past results, shallow soils, shallow casing
    - Agricultural areas
- sample four times
  - **August 08:** summer low flow
  - **November 08:** low flow (no recharge events)
  - **February 09:** melting event
  - **March 09:** melting event
- collected by reps from county agencies & DNR

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Methods

- bacteria → UW-Oshkosh Environmental Microbiology Lab
- **coliform:** surface water, soil, and fecal waste
  - safe levels: 0 coliform/100mL
- ***Escherichia coli:*** fecal coliform
  - conclusive evidence of fecal contamination
- **enterococci*:*** bacteria found in fecal waste
  - may be more reliable indicator than E. coli

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Methods

- nitrate and EDCs analyzed in UWGB labs
  - **nitrate**: measured using Lachat QuickChem 8500 Flow Injection Analysis System
  - **EDCs**: organics extracted with a carbon disk
    - within 24 hours
    - Wisconsin State Lab of Hygiene's protocol for the extraction of organic compounds from water

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E screen assay

- Used to assess estrogenicity of samples
- Greater estrogenicity → faster proliferation of MCF7-BOS breast cancer cells
- Standard curve range: 0.05 pM – 10,000 pM 17β estradiol
- Samples:
  - 100-, 200-, 400-, 800-, 1600-fold dilutions
  - Spikes, ICI antagonist, and blank samples included in each 24 well plate

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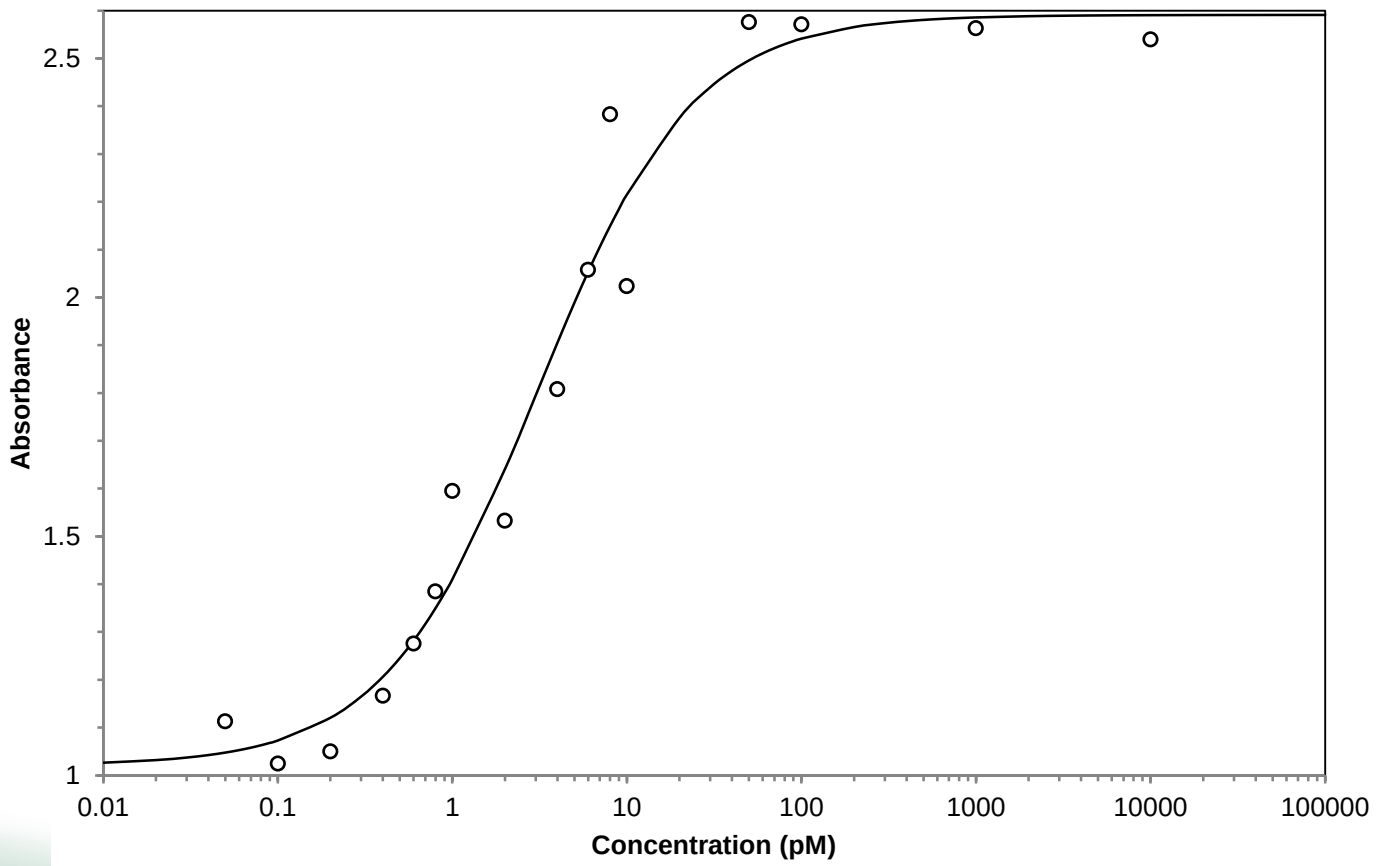

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Table 1. Percentage of groundwater wells falling in different nitrate concentration ranges during each sampling period.

Sampling Period	No. Wells Sampled	Concentration (mg/L NO_3-N)			
		0 – 2	2 – 5	5 – 10	> 10
1	40	17.5 %	7.5 %	20.0 %	55.0 %
2	37	21.6 %	8.1 %	21.6 %	48.7 %
3	39	18.0 %	12.8 %	18.0 %	51.3 %
4	37	11.1 %	33.3 %	33.3 %	22.2 %

Table 2: Percentage of groundwater wells with unsafe levels of coliform, enterococci, and *E. coli* during each sampling period.

Sampling Period	Coliform	E. Coli	Enterococci
1	62.5 %	12.5 %	27.5 %
2	40.5 %	2.7 %	10.8 %
3	59.0 %	7.7 %	29.7 %
4	64.9 %	27.0 %	46.0 %

Table 3: Percentage of sampled groundwater wells with detectable estradiol equivalents (EEq) in the E-screen assay during each sampling period.

Sampling Period	EEq Detections [a]
1	50.0 %
2	27.0 %
3	13.9 %
4	5.4 %

[a] Limit of sensitivity = 1 pM EEq in sample extracts

Table 4: Summary of significant differences in average concentrations between sampling periods for susceptible well water samples measured in this study.

Parameter	Sampling Periods					
	1 and 2	1 and 3	1 and 4	2 and 3	2 and 4	3 and 4
Nitrate	-	-	1 > 4	-	-	-
Conductivity	-	-	1 > 4	-	2 > 4	-
Coliform	-	-	4 > 1	3 > 2	4 > 2	4 > 3
Enterococci	1 > 2	-	-	3 > 2	4 > 2	4 > 3
E. coli	-	-	4 > 1	-	4 > 2	-
E-screen	-	1 > 3	1 > 4	-	2 > 4	-

Health Implications

- No public health standard for Eeq
  - EPA screening program
  - Nano-molar range
  - Conservative value might be 1 ng/L
- One well tested above 1 ng/L in the E-screen
  - B12-2: 3.5 ng/L
  - BD in other periods
- Most wells tested well below this range
  - Anti-estrogenic effects?
  - Toxicity?

Conclusions

- Groundwater contamination with nitrate, bacteria and EDCs is a common problem in karst areas of NEW
- EDC contamination was greater during the months of August and November
- Bacterial contamination was greatest during the months of February and March
- No correlation exists between water quality parameters

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