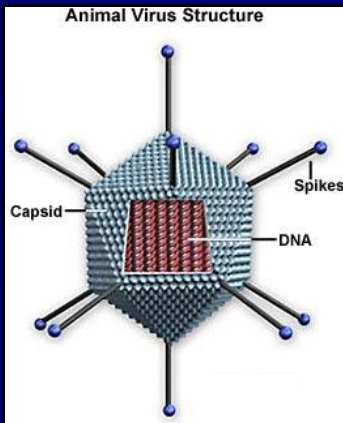


# ***SURVIVAL OF CONTAMINANT MICROORGANISMS IN MODEL ASR CONDITIONS FOR FLORIDA***

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# Research Objectives

- Bench-scale survival studies of several groups of microorganisms in representative surface water and ground water sources with potential for or in active utilization for ASR systems, focusing on temperature and background microbial community effects (pasteurized subsamples)
- Saturated limestone study to investigate impact of solid media and no aeration on survival of select organisms

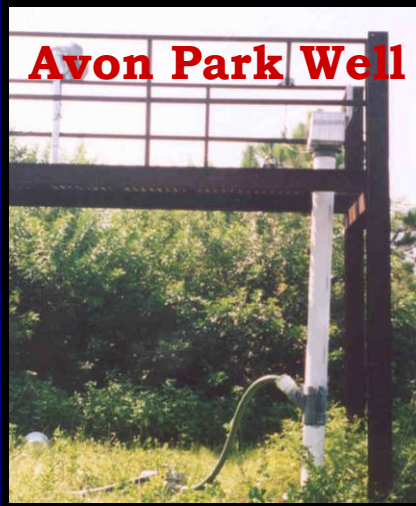
# Survival in Representative ASR Source and Receiving Water

- Desired specific description of survival behavior in waters typical of those for ASR projects

**Bill Evers Reservoir**



**Avon Park Well**



**Clear Lake Reservoir**

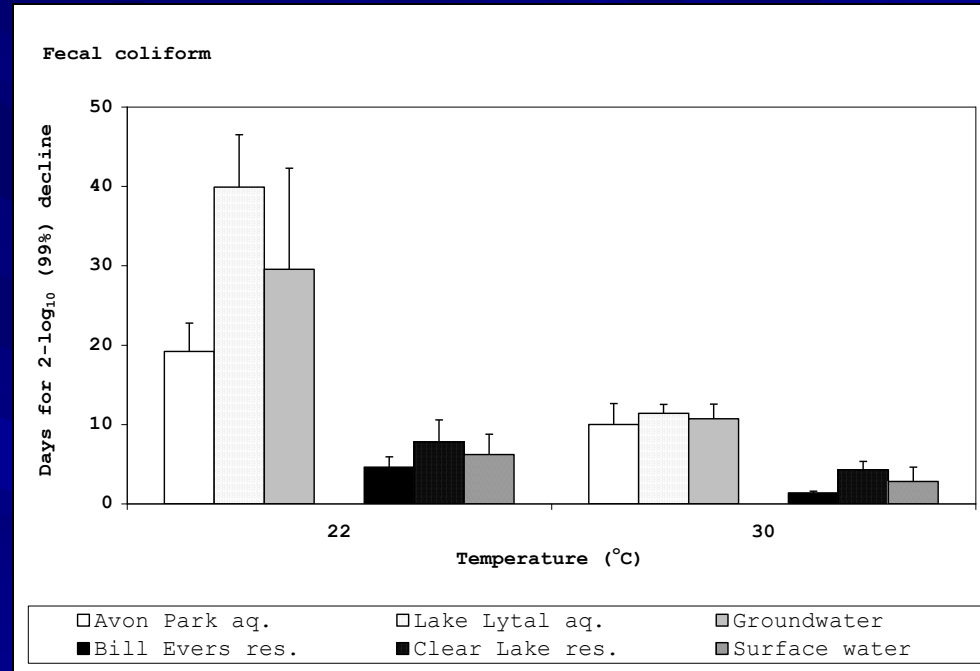


**Lake Lytal  
Park Well**



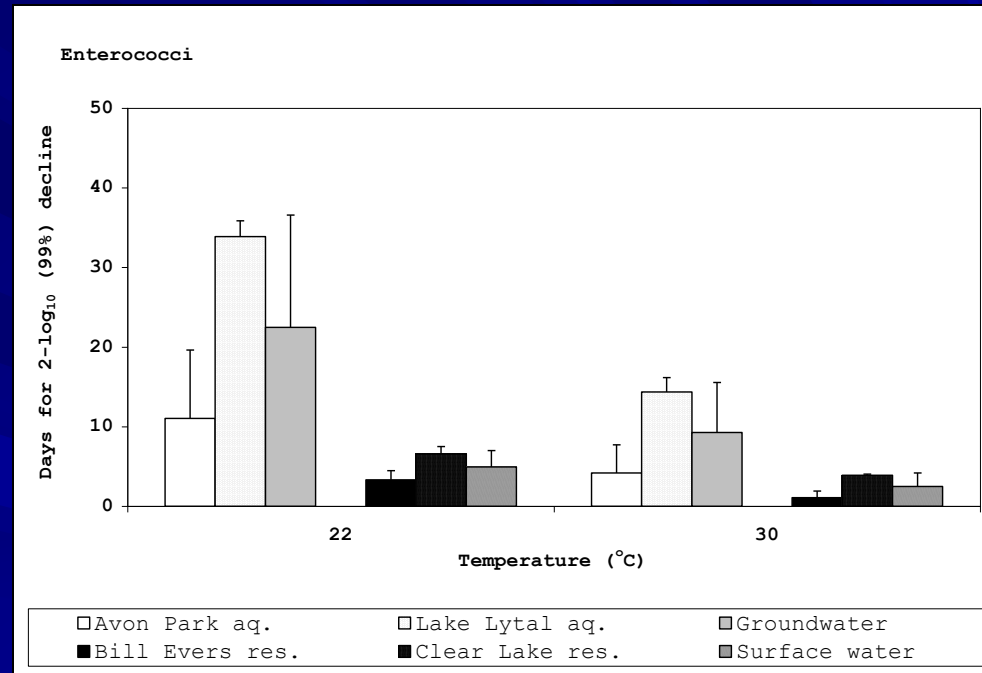
# Fecal Coliform Survival

- Relative effects:
  - statistically significant
    - increasing inactivation with temperature 5° - 30°
    - longer survival in pasteurized
    - more rapid inactivation in surface water, mostly at higher temperature
    - significance of temperature varied with water type in independent analyses



# Enterococci Survival

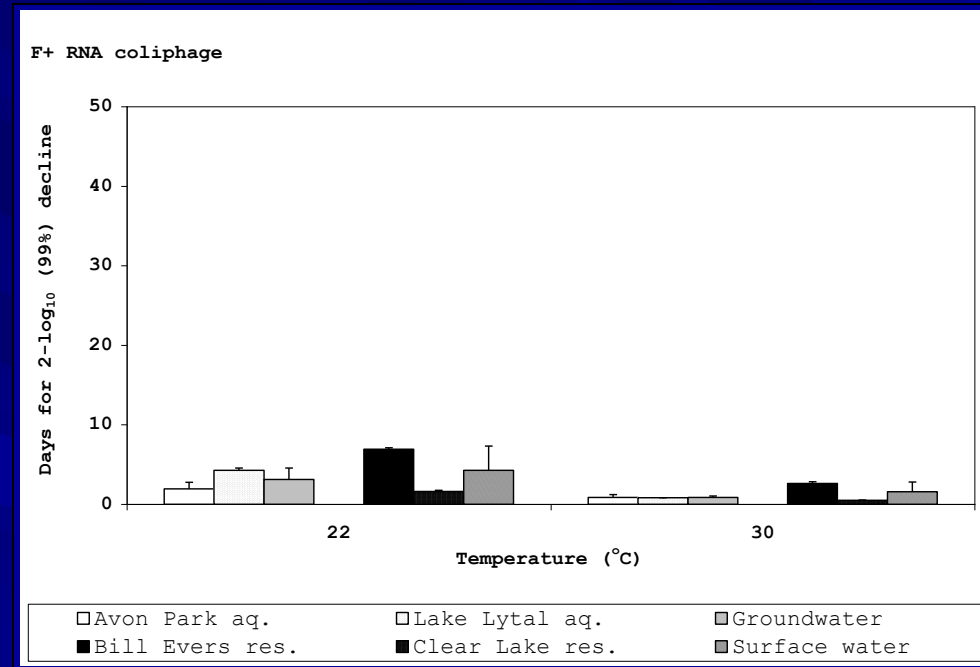
- Statistical analysis of inactivation revealed:
  - significant increase with temperature, reduced inactivation in pasteurized water, faster inactivation in raw surface water vs. raw ground water
  - interaction of water type, temperature, and pasteurization



# F+ RNA Coliphage Survival

## ■ Observations of survival behavior included:

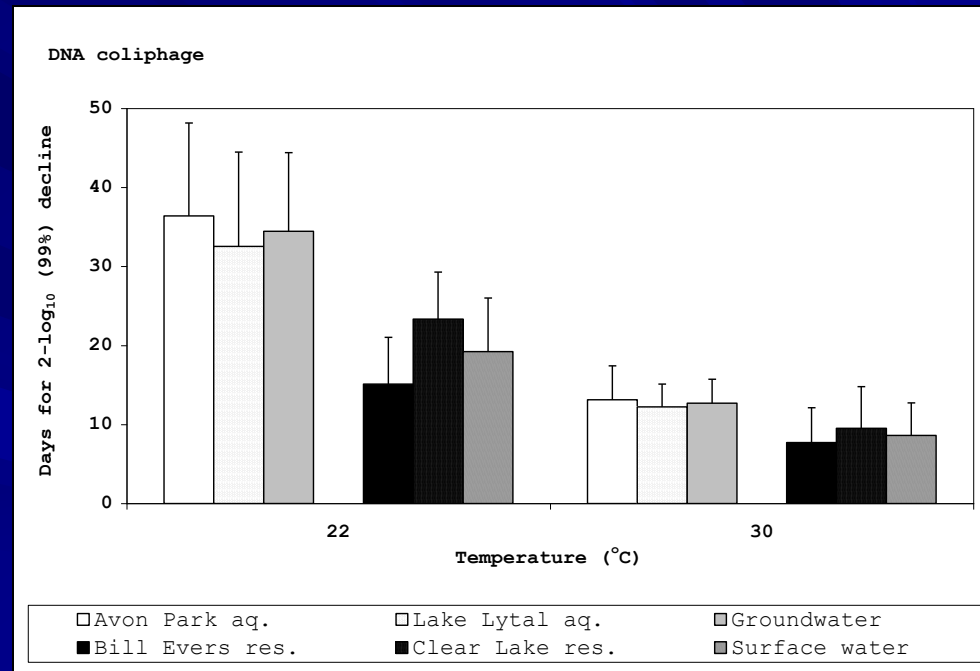
- temperature significantly affected inactivation
- Treatment and water type effects due to differences only at low temperature
- uniformly rapid inactivation at environmental temps.



# DNA Coliphage Survival

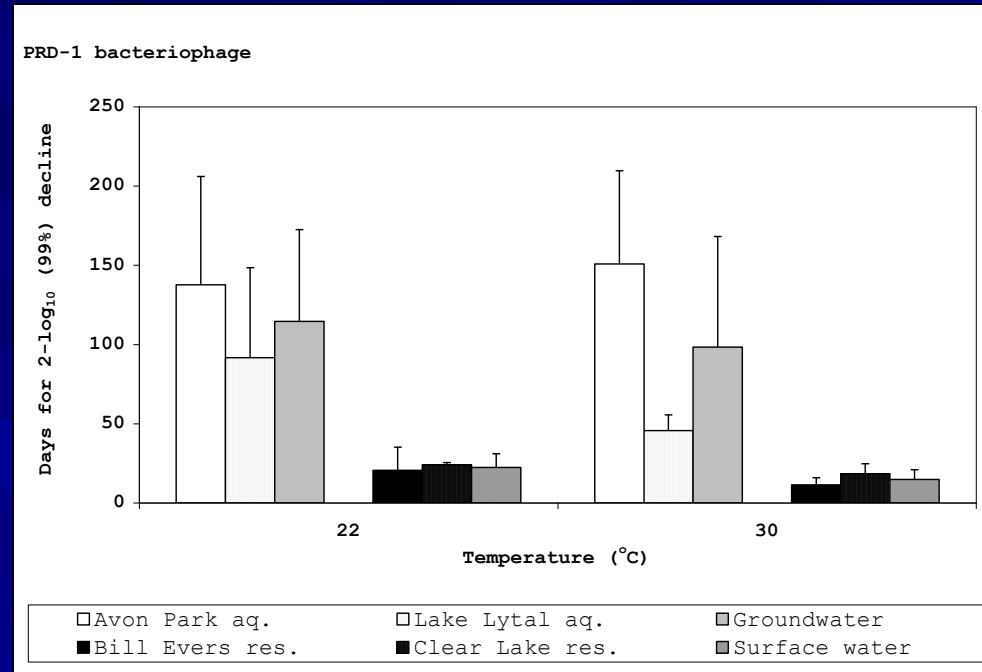
■ Significant trends in inactivation were:

- increasing inactivation with temperature, and in raw
- pasteurization had more of an effect in surface water, but affected both with less rapid decline in pasteurized (not significant for ground water separately)
- water type effects only significant due to differences from pasteur-ized samples



# PRD-1 Survival

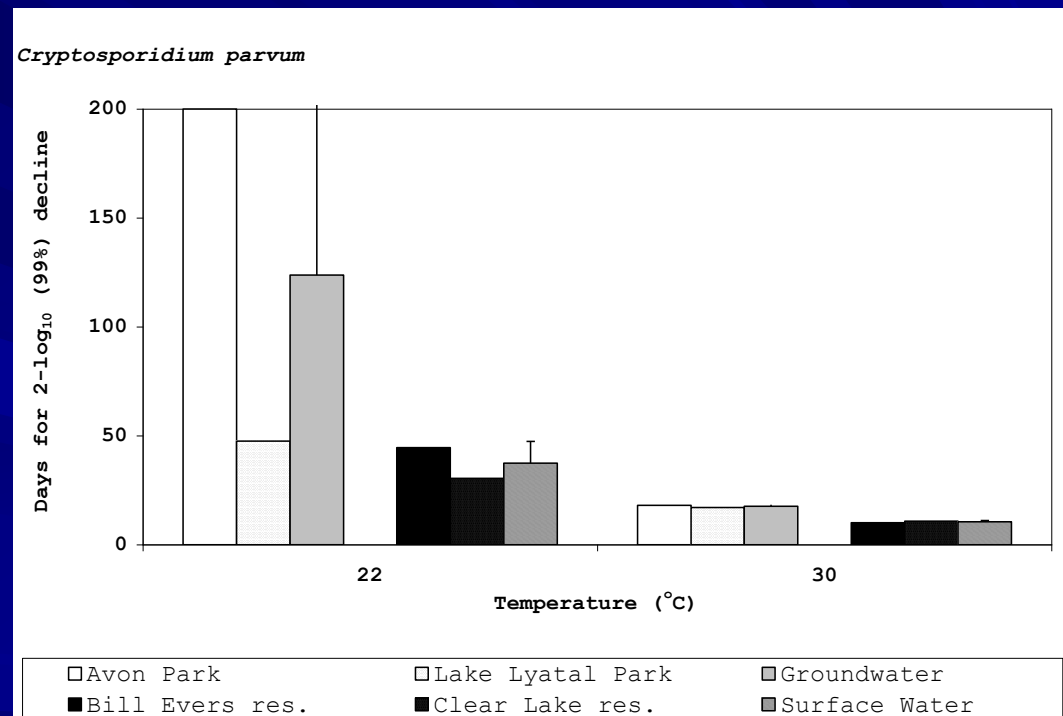
- Slower inactivation than other organisms on average
- Statistically-significant trends were:
  - increased inactivation with temperature and in surface water
  - in raw water, temperature effect interaction with water type (surface water affected more)
- Pasteurization not significant



# *Cryptosporidium* Survival

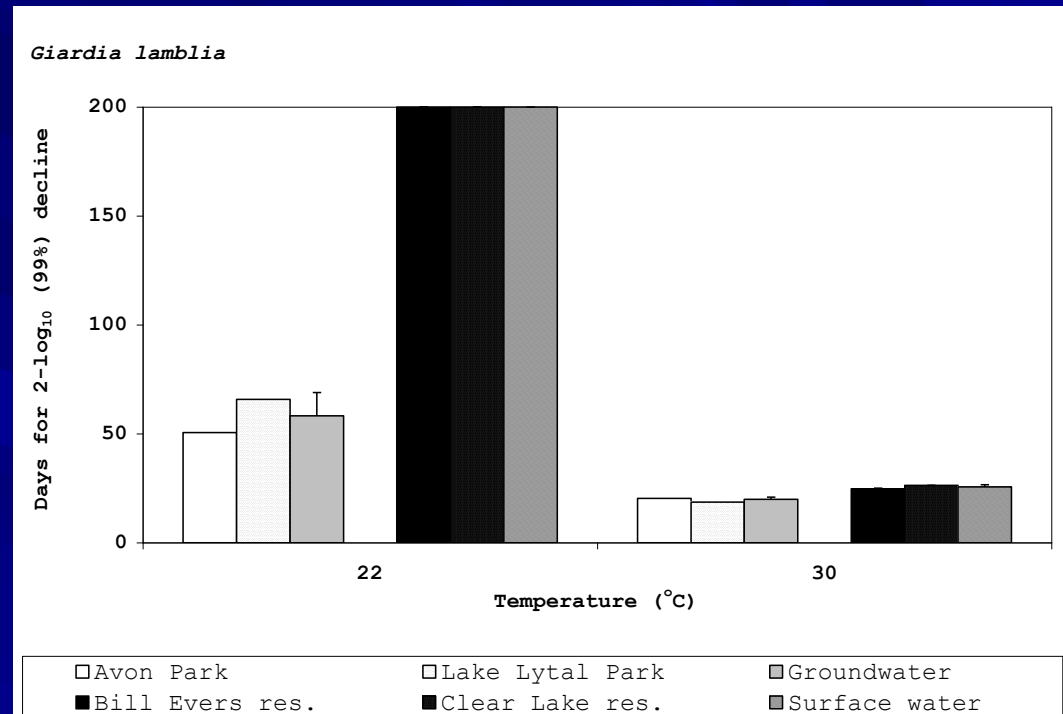
■ Pasteurized samples not used; temperature and water type comparison

- temp., water type, and interaction significant
- faster inactivation at higher temperature
- more rapid inactivation in surface water, less difference at higher temperature



# *Giardia* Survival

- In temperature and water type comparison:
  - significant temperature effect
  - More rapid inactivation in ground water than surface water
  - Water type differences most pronounced at 22°, similar at 5° and 30° C



# Important Facets of Survival in ASR Water Samples

- Higher temperature consistently increased inactivation, pasteurization decreased inactivation, and inactivation more rapid in surface water for bacteria, virus, and *Cryptosporidium*
- Possible antagonistic effects of native microbial populations, more important in surface water? (enterococci and DNA coliphage)
  - metabolic activity of surface water organisms
  - oxidative and pressure conditions: impacts on contaminant and native organisms?

# Comparisons of Organism Populations

- At temperatures of Floridan aquifer system ( $22^{\circ} - 30^{\circ}$  C), fecal coliform and enterococci fairly similar
  - fecal coliform 2-log (99%) decline in 2-6 weeks ground, 1-2 weeks surface; enterococci slightly less at 1-5 weeks ground, 1 week surface
- DNA coliphage much hardier than RNA coliphage
  - F+ RNA coliphage  $< 1$  week in both types; DNA coliphage 2 – 6 weeks in both types
- PRD-1 showed slower decline:
  - 6 months predicted in Avon Park at both temps, 2 – 3  $\frac{1}{2}$  months Lake Lytal Park; 2 -3 weeks in surface water

# Comparison of Parasite Inactivation Rates

## ■ In ground water samples:

- *Crypto* much slower than bacteria in Avon Park at 22° C (1/100<sup>th</sup>), only slightly slower in Lake Lytal Park at 22° C and both samples at 30° C
- *Giardia* rates about 1/2 of bacteria at 22° and 30° C

## ■ In surface water samples:

- *Giardia* rates very slow (no inactivation) at 22° C, at 30° C rates about 1/10 to 1/5 of bacteria
- *Crypto* rates about 1/10 of bacteria at 22° C, and about 1/2 to 1/3 of bacteria at 30° C

# Saturated Limestone Study

- Attachment often key in microorganism behavior, air-water-solid interface may be important
- Ocala limestone used to provide “aquifer” matrix
- Water sources from same representative ASR sites used, only higher temperatures of Floridan Aquifer system

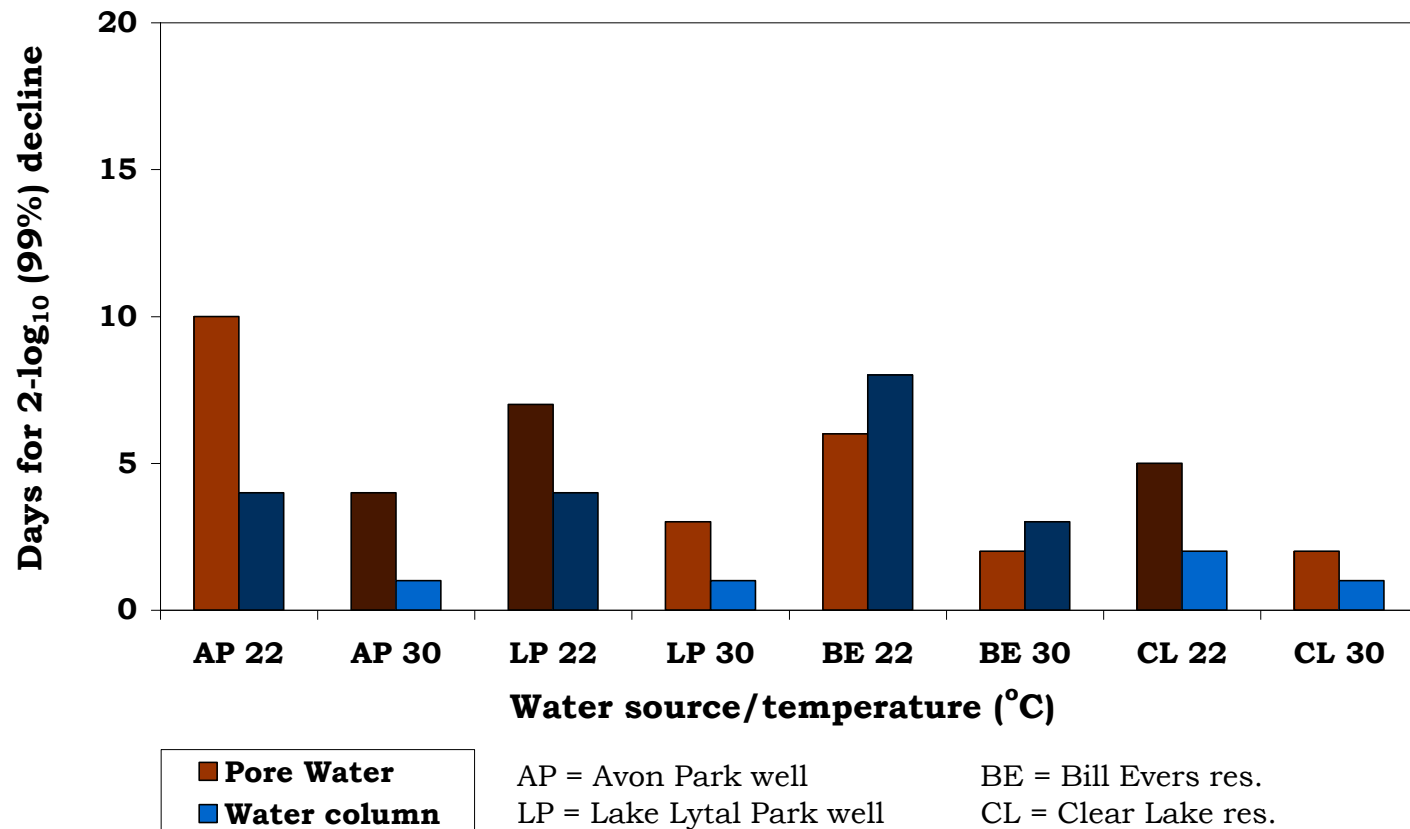
# Pore Water Study Methods

- Ocala limestone from Zephyr mine
- Crushed, washed and conditioned in UFA water
- Organisms only fecal coliform, enterococci, F+ RNA coliphage; seeded into water, limestone added
- Sampling of entire pore-water volume (discrete samples)



# Results Comparison – Pore Water to Water Column

## F+ RNA coliphage



# Pore Water Study Findings

- Definite increase of survival potential for bacteria in surface water saturated limestone
- Difference in ground water not clear for bacteria
- F+ RNA coliphage still most rapidly inactivated
- Mechanisms?
  - Aerobic metabolism of surface water organisms
  - protection due to attachment
- Important first look at limestone saturated with these water types

# Concluding Remarks

- Die-off does occur, temperature effects on survival mean likely more rapid decline than in colder areas
- Differences with pasteurized samples indicate native microbes may reduce survival of contaminant enteric bacteria and viruses
  - more important in surface water?
- Intestinal parasites may outlive bacterial indicators substantially



# Concluding Remarks - 2

- Environmental differences between microcosms and aquifer
  - aeration, redox, pressure, mineral surfaces, chemical differences: all may alter survival behavior of contaminant and native microbes
- Inactivation reduced in saturated limestone pore-water
  - Why?
  - Recommend larger scale models, examine effects of subsurface on surface water microbes
- Sound science necessary to keep pace with implementation



# Acknowledgements

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