FATE AND TRANSPORT OF PATHOGENS WITHIN LAND APPLIED BIOSOLIDS

Ian L. Pepper
University of Arizona
Tucson, Arizona

PATHOGENS OF CONCERN

ORGANISM	EXAMPLE
Virus	**Hepatitis A**
Bacteria	***Salmonella typhi***
Protozoa	***Cryptosporidium parvum***

MAJOR ROUTES OF EXPOSURE FROM LAND APPLICATION OF BIOSOLIDS

AIR

- Transport on bioaerosols
- Transport on colloidal particles
- Very little data available

SOIL

- Transport via solution phases
- Large data base for pure culture transport (no biosolids)
- Very little data on transport of pathogens originally contained in biosolids

BIOAEROSOLS

- ***Bioaerosol*** **defined as collection of aerosolized biological particles (microbes)**
- **Vary in size from 0.02–100 💻m in diameter**
- **Composition, size and concentration of microbial populations within the bioaerosol varies with source of microbes and environmental conditions**
- **Can be associated with water or soil colloidal particles**
- **Virus and bacteria have greatest potential for transport—protozoans too large**
- **Fungal spores and hyphae can also be dispersed as bioaerosols**

FACTORS AFFECTING VIABILITY OF BIOAEROSOLS (Conventional Wisdom)

- Duration in air ie. high wind speed can enhance transport but reduce viability
- Gram negative bacteria survive longer at low relative humidity
- Gram positive bacteria survive longer at higher relative humidity
- Higher temperatures reduce viability
- UV radiation inactivates pathogens

LAUNCHING OF BIOAEROSOLS FROM BIOSOLIDS

- *Point source*—biosolid pile
- *Area source*—land applied field
- Biosolids considered to be *continuous source* as opposed to *instantaneous point source*

FACTORS INFLUENCING BIOAEROSOL TRANSPORT

- **Transport 🖰 size of bioaerosol ✆**
- **Transport 🖰 as wind speed 🖰**
- **Transport occurs via thermal gradients from regions of warmer temperatures to lower temperatures**

MODELING TRANSPORT OF BIOAEROSOLS FROM LAND APPLIED BIOSOLIDS

- Scale of transport
  - –submicroscale: 10 minutes, 100 m
  - –microscale: up to 1 hour, 100m to 1 km
- Point source modeling
  (Pasquill, 1961. Meteorol. Mag. 90:33–49)
- Area source modeling
  (Parker et al., 1977. J. Water Pollut. Control Fed. 49:2359–2365)

STUDIES ON BIOAEROSOLS FROM LAND APPLIED BIOSOLIDS

- Overall very little information available
- Three major studies
  - Dowd/Pillai (Texas)
  - Pepper/Gerba* (Arizona/California)
  - Millner/Walker* (East Coast)

*Still in progress

BIOAEROSOLS: DOWD/PILLAI

Bioaerosol Transport Modeling and Risk Assessment in Relation to Biosolid Placement

J. Environ. Qual. 29:343–348
Year: 2000
Authors: Dowd, Gerba, Pepper, Pillai

RISK FOR VIRAL INFECTIONS

SOURCE	DISTANCE (m)	WIND SPEED (Low or High)	DURATION OF EXPOSURE (hours)	RISK (%)
Point	100	Low	1	3
Area	100	Low	1	6

Populations 10,000 m away essentially zero risk

BIOAEROSOLS: DOWD/PILLAI

- Deliberately looked for worse case scenario
- Deliberately used conservative approach ie. over estimate risk
- No human enteric pathogens actually measured other than *Salmonella*
- Data only applicable to that study

This paper has been used inappropriately to overstate the potential for bioaerosol production from land application.

BIOAEROSOLS: MILLNER/WALKER

- Focused on chemical odors
- Evaluating odors per se as potential health hazard
- Are not looking at biologicals in detail
- Study still underway

The University of Arizona National Science Foundation Water Quality Center

Director: ***Ian L. Pepper***

An Industry/University Cooperative Research Center (I/UCRC)

BIOAEROSOL STUDY: PEPPER/GERBA, 2002

CURRENT BIOAEROSOL RESEARCH

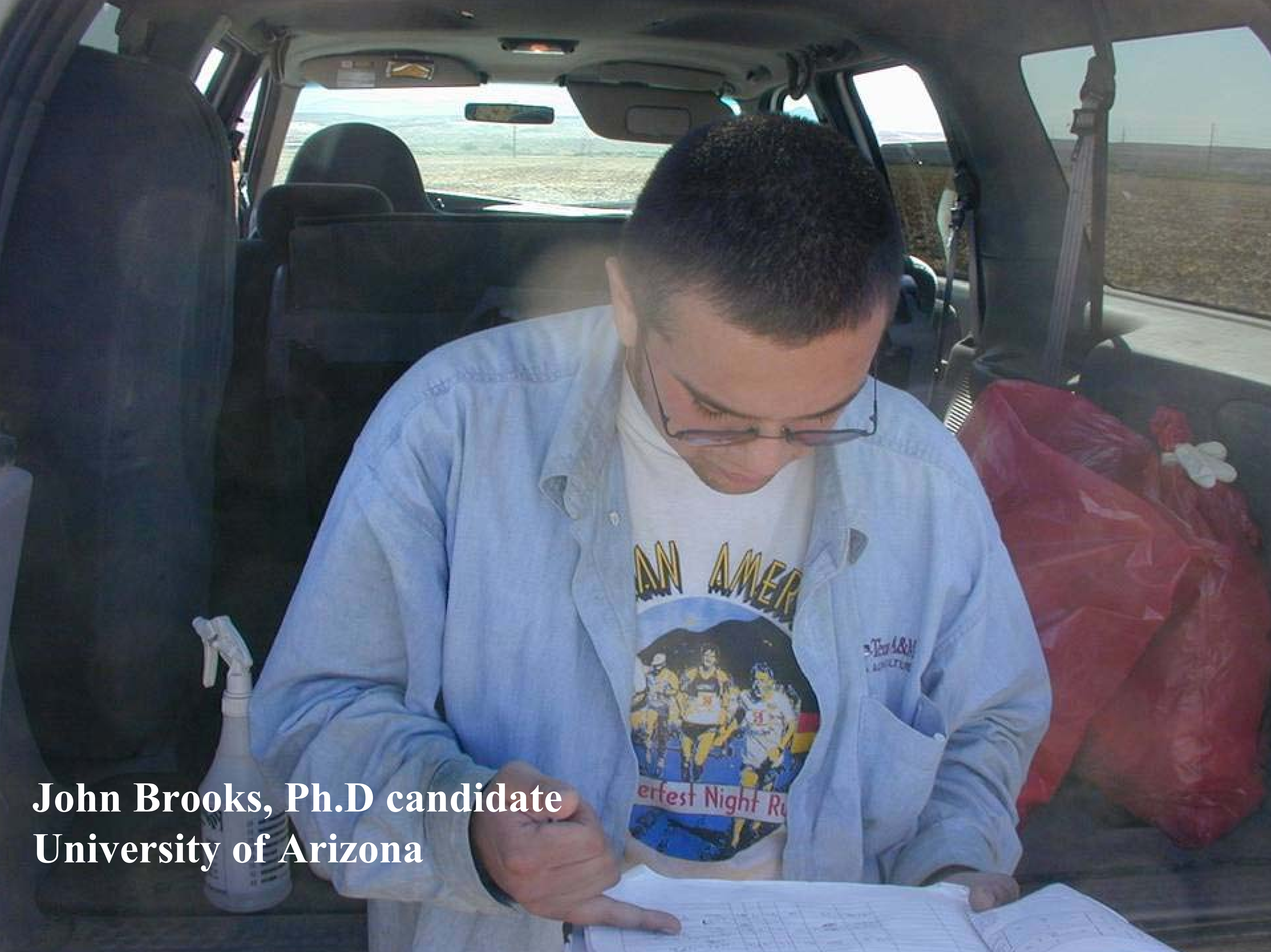
- Two Ph.D. candidates—
  John Brooks and Ben Tanner
- P.I.'s—Ian Pepper and Charles Gerba
- Over 300 samples analyzed
- Odors clearly a real problem

BIOAEROSOL RESEARCH

- **Samples analysed for heterotrophic plate count bacteria, total coliforms, *E. coli*, *Salmonella*, enteroviruses phage, *Clostridium perfringens*, *Aspergillus* spp.**

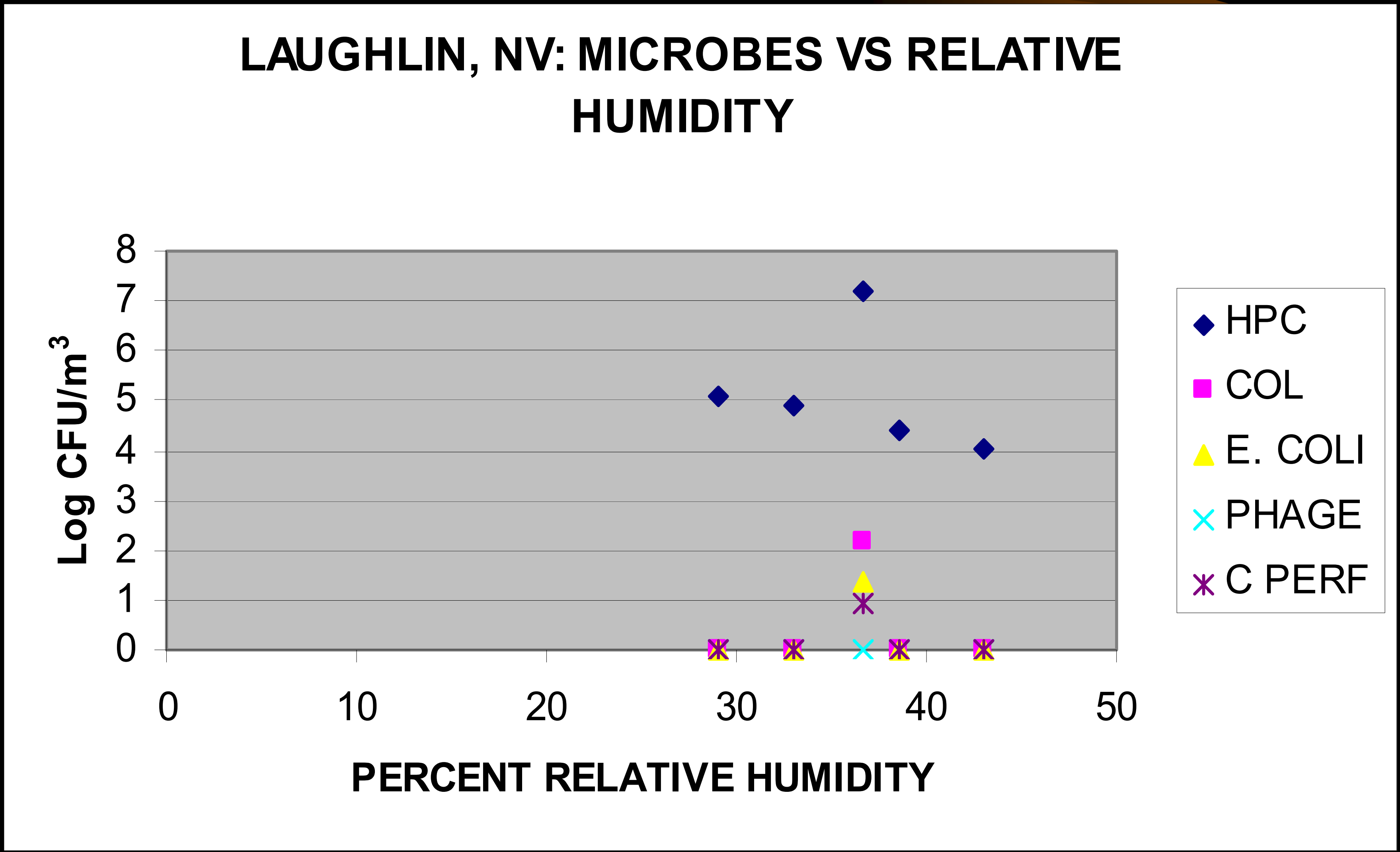

CRAFTSMAN


5000

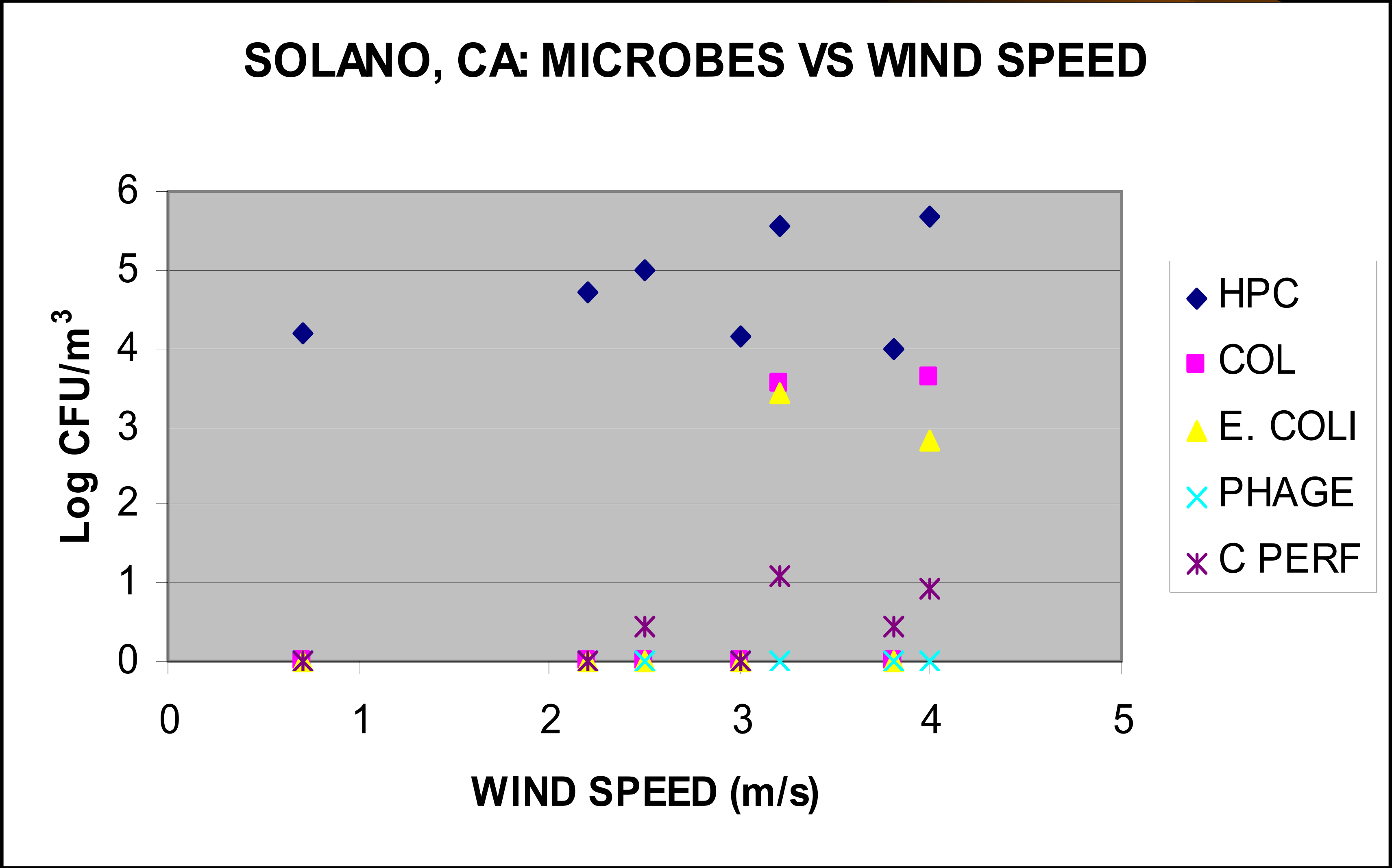


John Brooks, Ph.D candidate
University of Arizona

Example of Research Data



Example of Research Data



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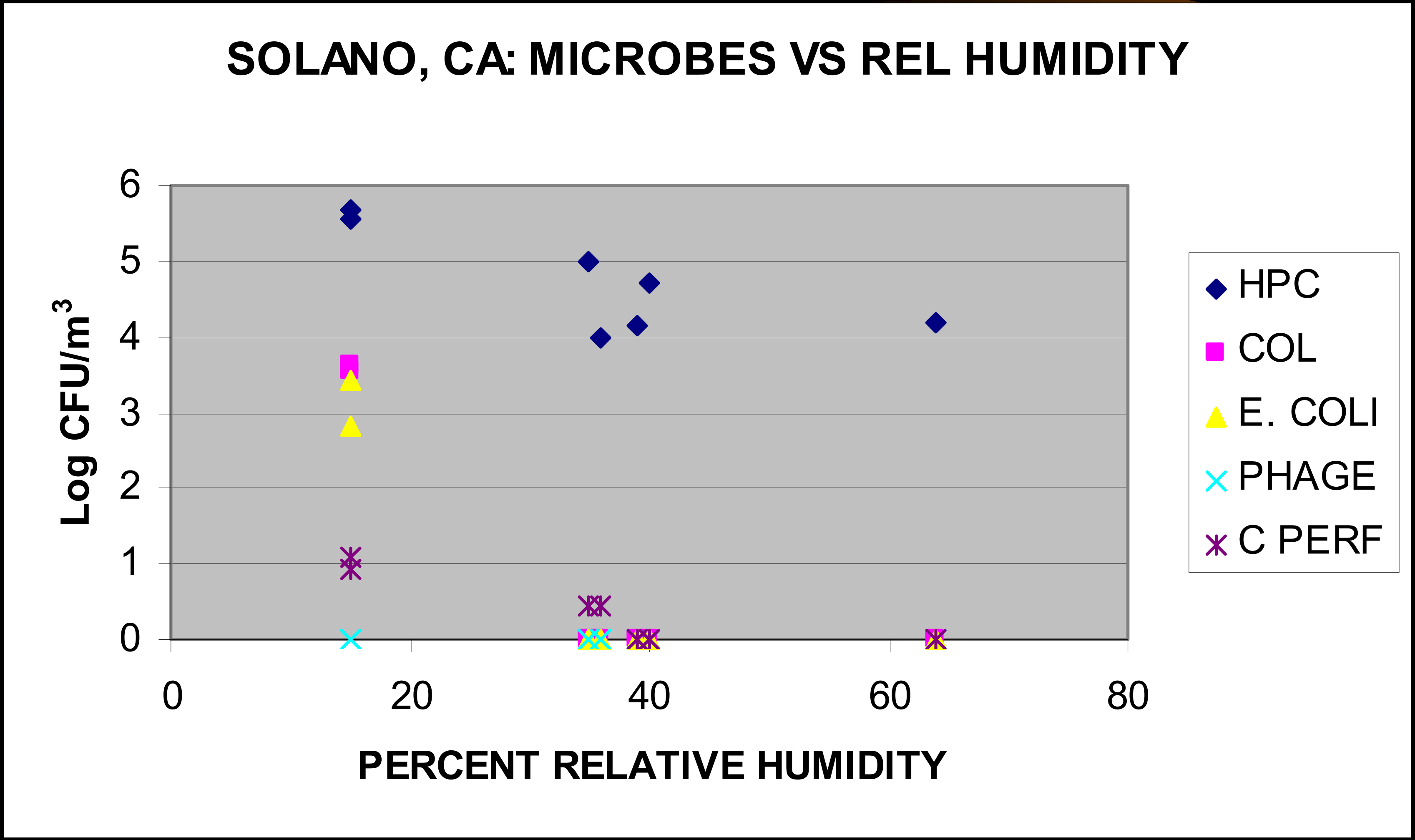


RESULTS

- **Bacteria counts from tractor operation (no biosolids) 10,000/ cubic meter of air**
- **Coliform bacteria (indicators not pathogens) found occasionally**
- ***E. coli* found occasionally**
- ***Salmonella* not found**
- **Phage not found**
- ***Staphylococcus aureus* not found**
- **Low concentration *Clostridium perfringens* found once during application**

DISCUSSION

- Dust responsible for HPC as well as biosolids
- No known enteric pathogens detected in the study
- Low concentrations of indicator organisms found (coliforms)
- Low windspeed reduces transport
- High temperatures reduces organism viability
- Low humidity reduces organism viability

BIOAEROSOLS: PEPPER/GERBA

- Study to continue for 1 more year
- Modeling of transport of viable microbes
- Modeling of environmental parameters influence on viability
- Risk assessment analysis based on predicted exposure scenarios and known dose response

SURVIVAL OF BIOSOLID-ASSOCIATED PATHOGENS IN SOIL

Numerous studies on survival of pathogens in soil.

- **Studies have looked at pathogen survival following land application of biosolids**
- **Laboratory batch studies—static**
- **Field studies—dynamic**

SURVIVAL OF BIOASOLID-ASSOCIATED PATHOGENS IN SOIL

	Survival Time		
Parameter	**Virus**	**Bacteria**	**Protozoa**
Temperature increasing	✎	✎	✎
Soil moisture decreasing	✎	✎	✎
Rate of desiccation increasing	✎	✎	✎
Clay content increasing			Not known
pH range of 6–8			

✎, decreasing survival time; +, increasing survival time

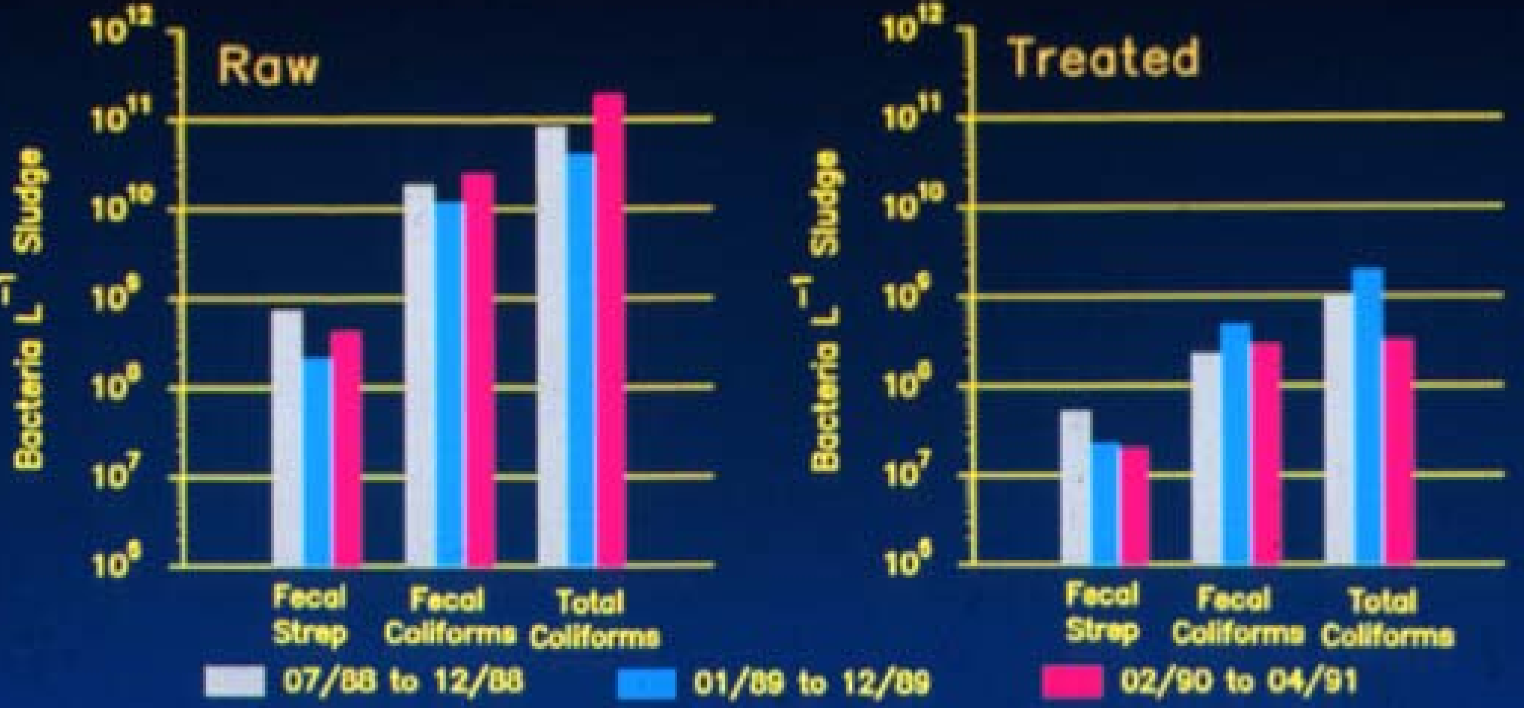
Source: Gerba et al., 1975; Straub et al., 1993a, 1993b, 1995, Jenkins et al., 1999

SURVIVAL OF BACTERIAL PATHOGENS IN BIOSOLID AMENDED SOIL

- ***Salmonella*** **and** ***E. coli*** **reported to survive up to 16 months**
- ***Shigella*** **survives for shorter duration**
- **Regrowth possible following rainfall events**

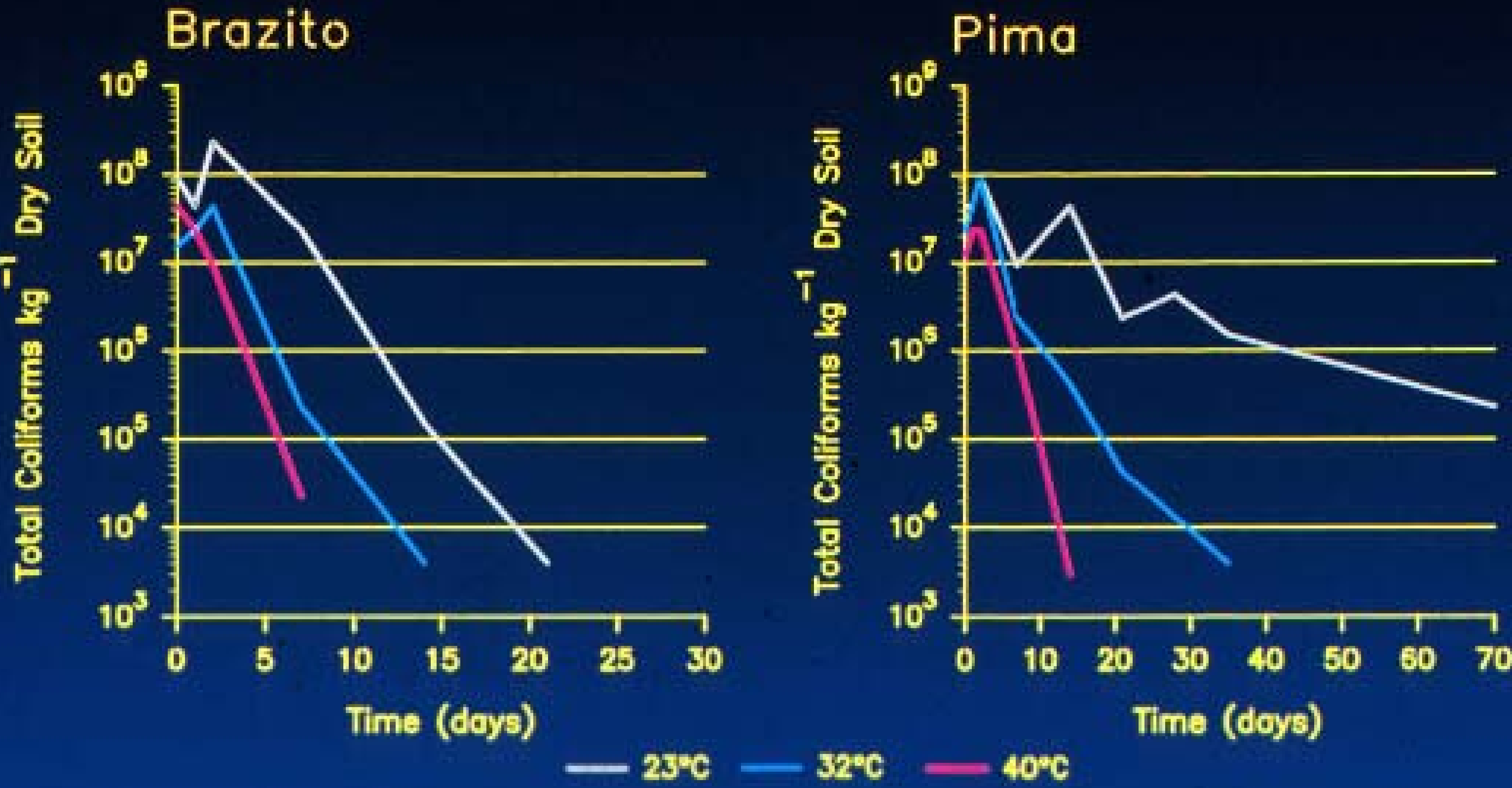
Pathogen Survival in Sewage Sludge

Raw vs. Treated Sludge



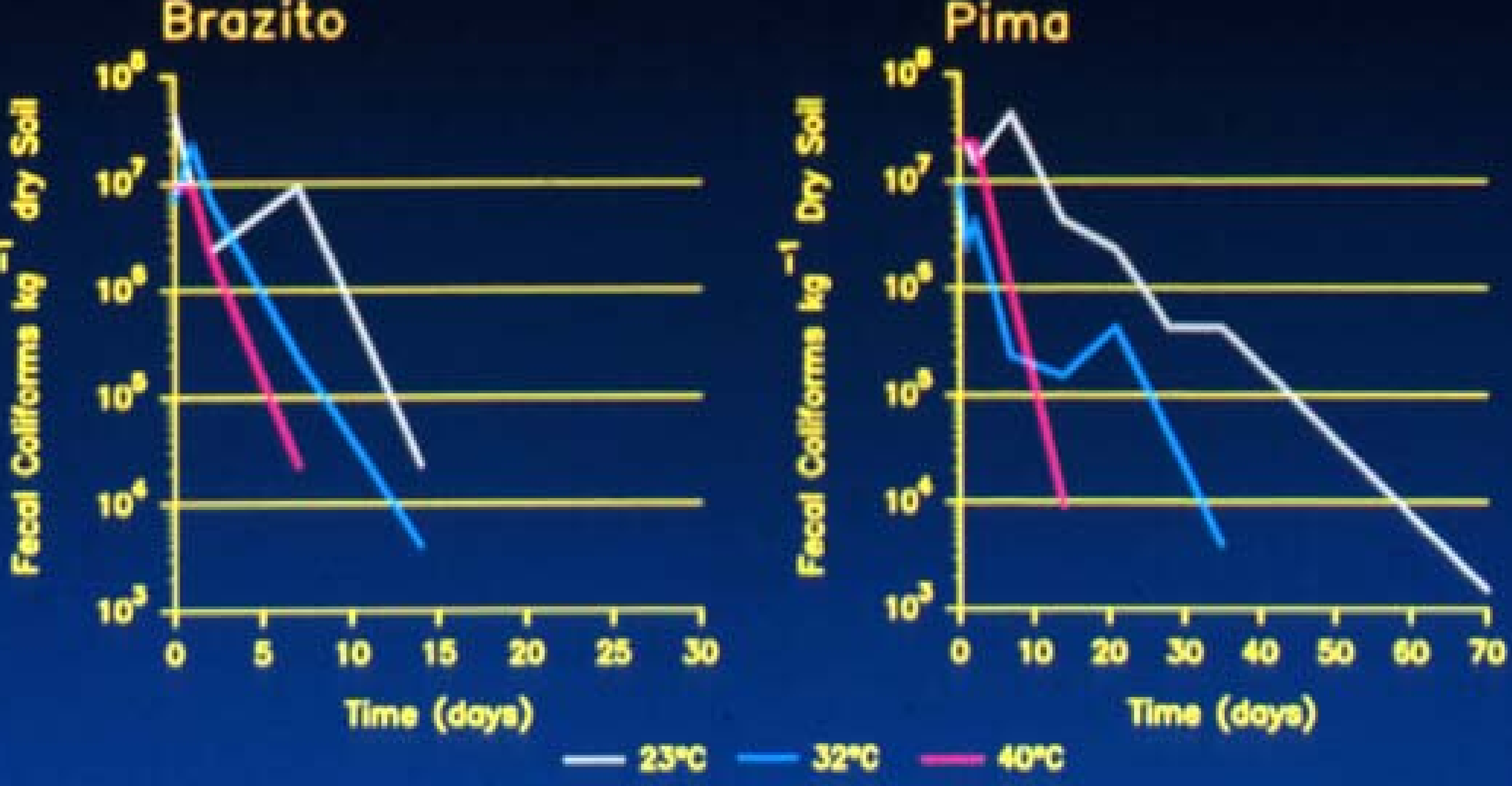
Survival of Total Coliforms

Laboratory Study



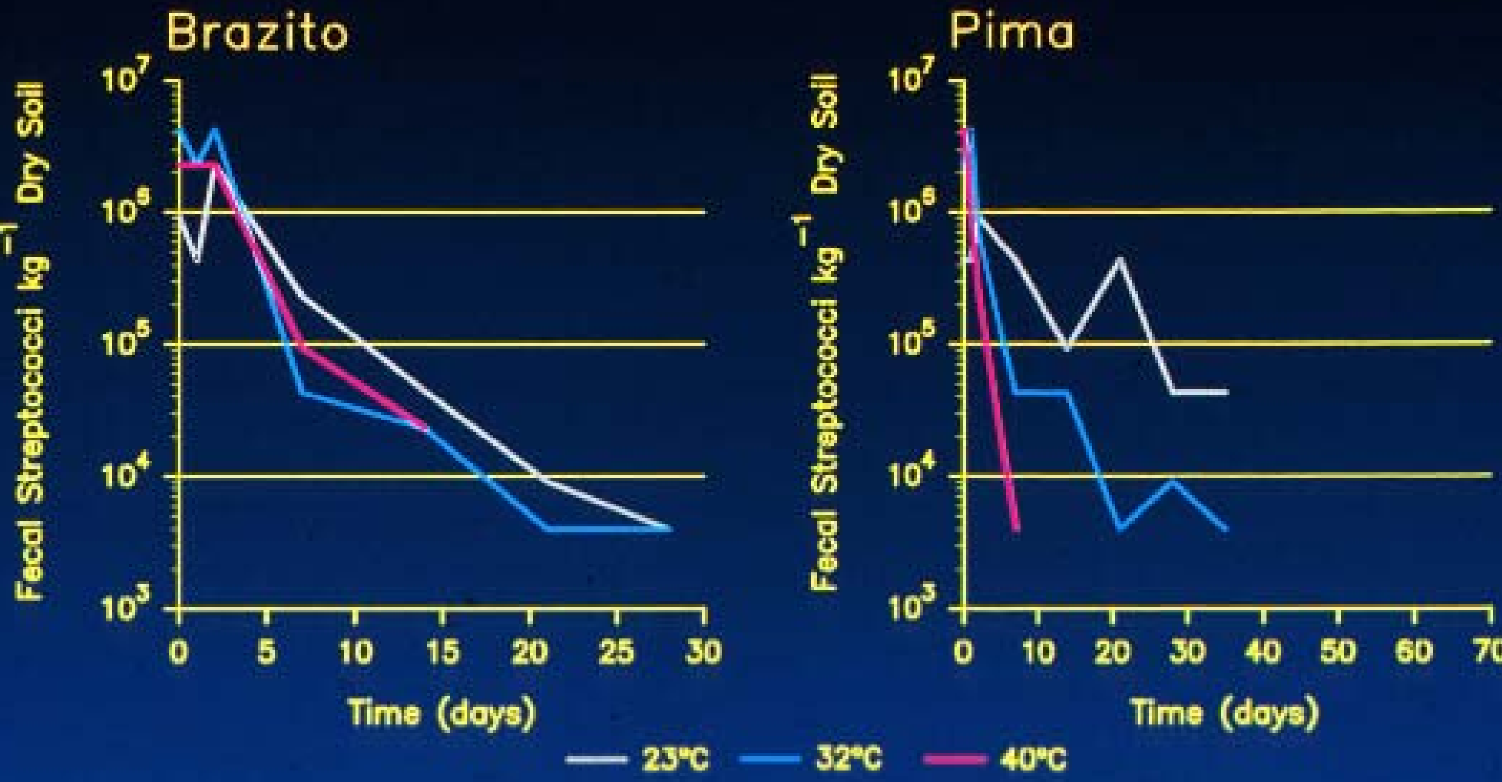
Survival of Fecal Coliforms

Laboratory Study



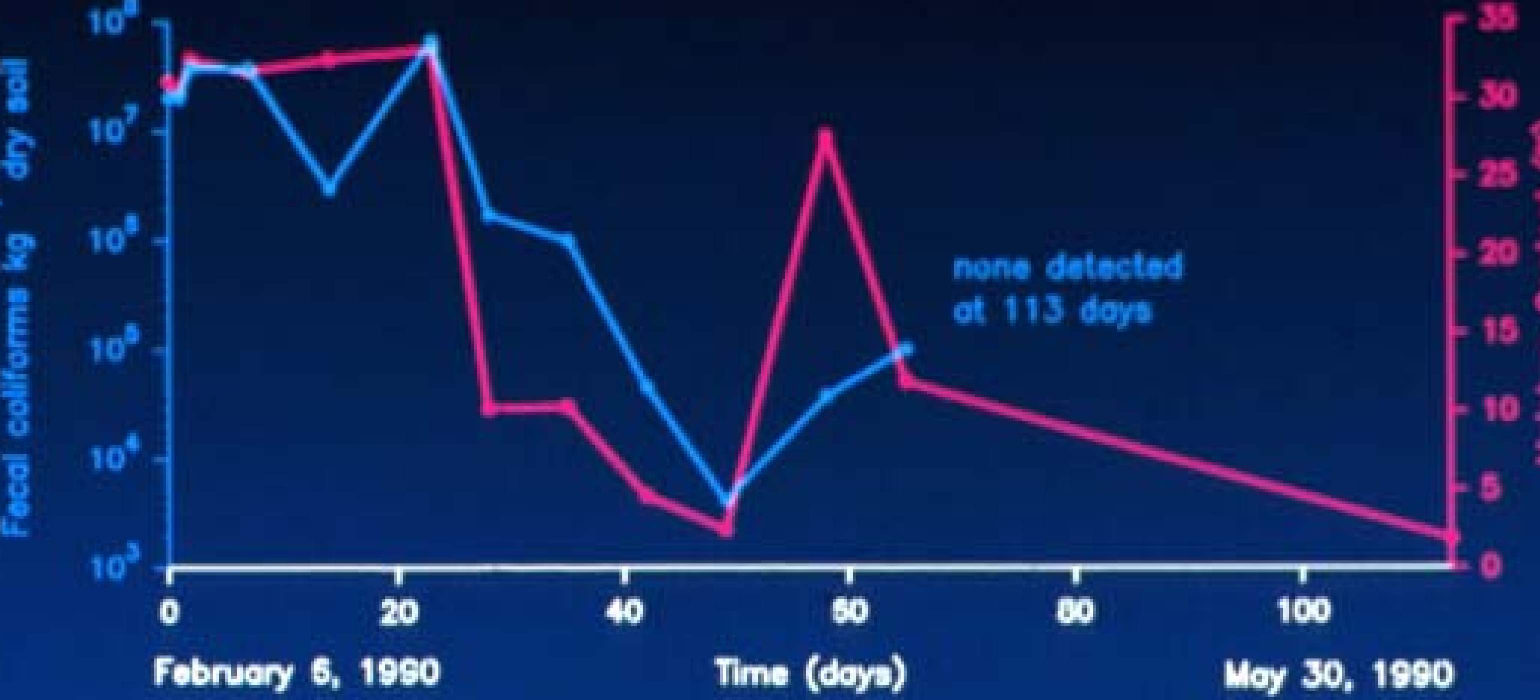
Survival of Fecal Streptococci

Laboratory Study



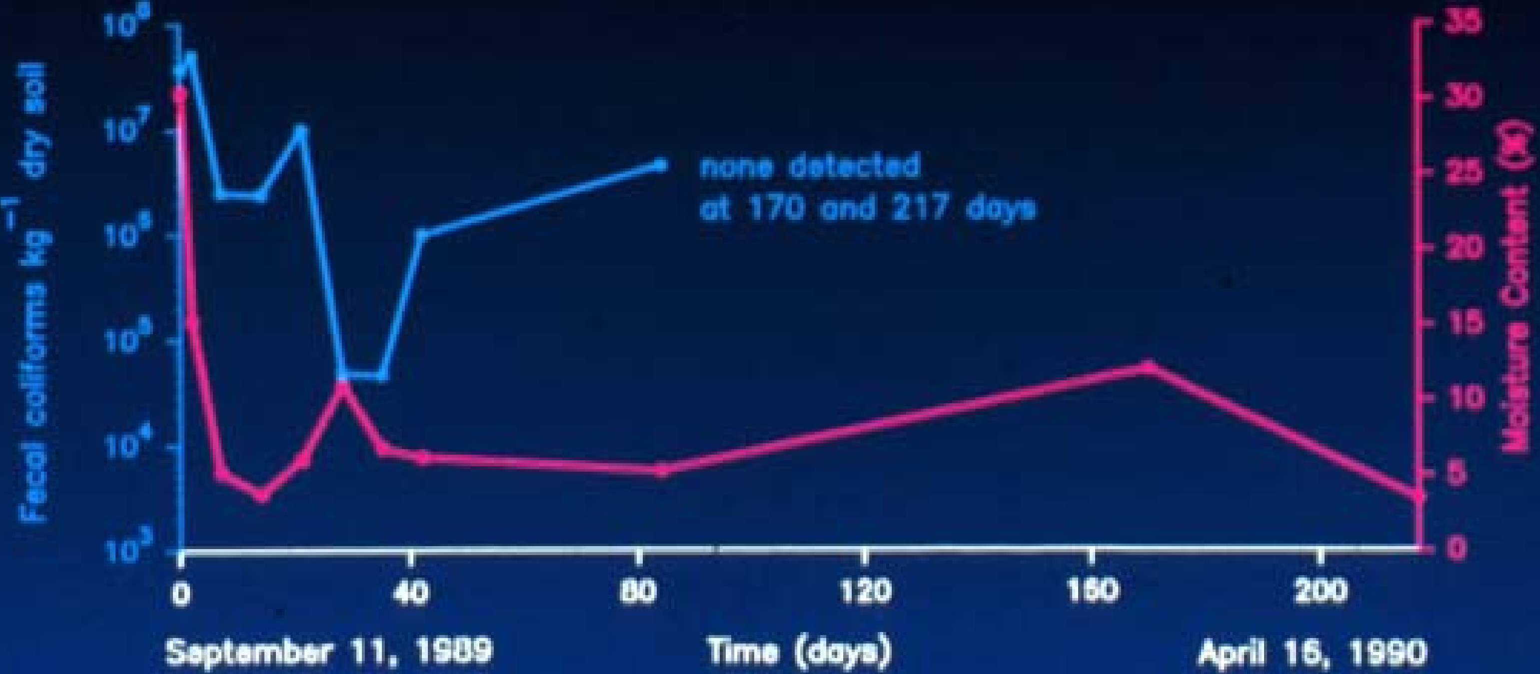
Survival of Fecal Coliforms - Marana Field

With Respect to Time and Soil Moisture Content



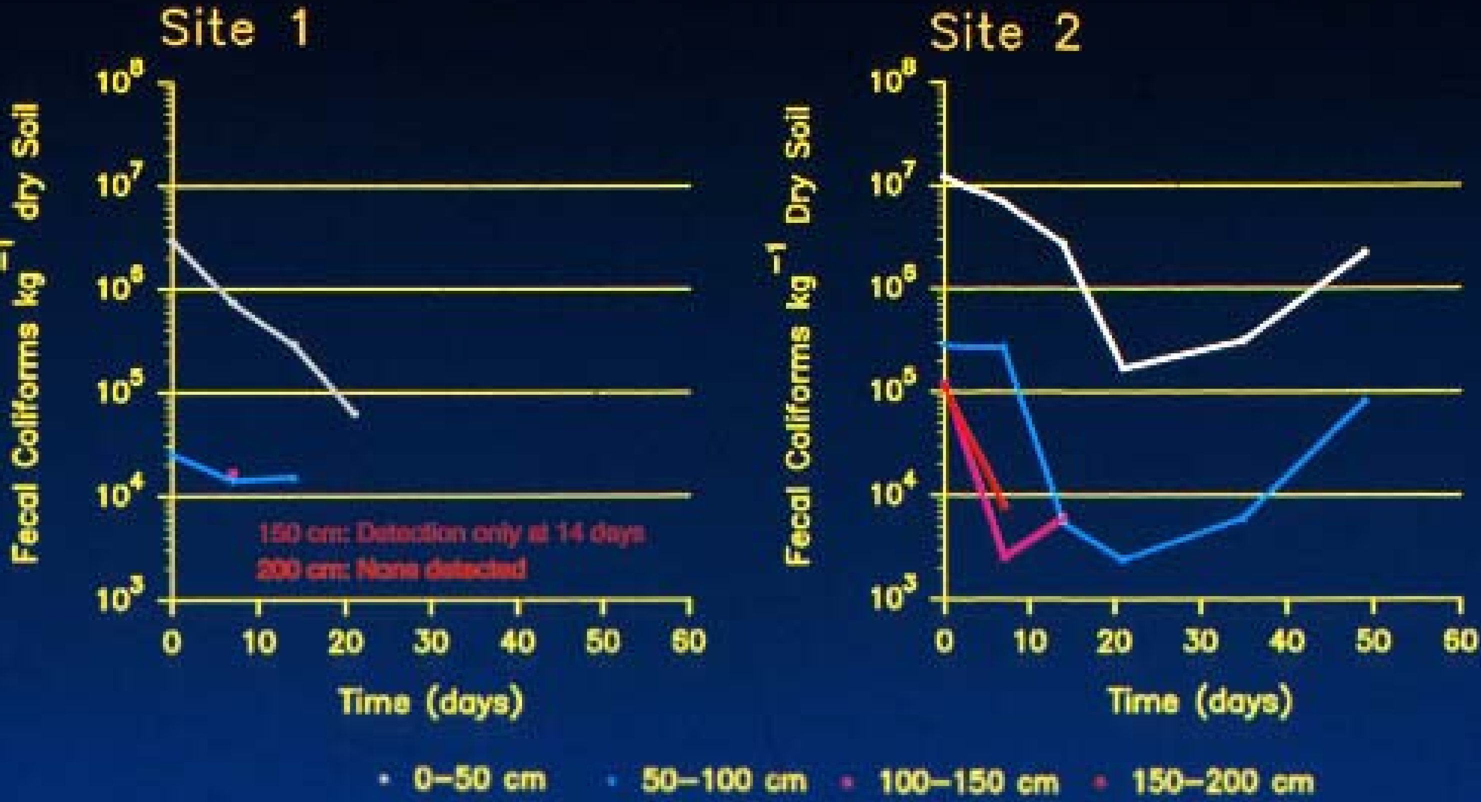
Survival of Fecal Coliforms - Avra Valley Field

With Respect to Time and Soil Moisture Content



Survival of Fecal Coliforms - Avra Valley Field

As a Function of Time and Depth



SURVIVAL OF PROTOZOAN PARASITES IN BIOSOLID AMENDED SOIL

- **Little work reported on *Giardia* and *Cryptosporidium***
- **Helminths persist longer than any other pathogen**
- **Ascaris eggs can be reported to survive several years in soil**
- **Size of protozoans (with exception of *Microsporidia*) preclude transport through soil**

SURVIVAL OF VIRAL PATHOGENS IN BIOSOLID AMENDED SOIL

- University of Arizona study, early 1990's
- Survival ✆ as temperature 🖰
- Survival better in clay loam than sandy loam
- Survival time of poliovirus detected via cell culture = 3–10 days depending on soil type, temperature and soil moisture
- survival time when detected via PCR = 3 months

METHODS FOR DETECTION OF VIRUS

- **Traditional approach—cell culture**
- **Molecular approach—PCR**
- **Latest approach—Integrated Cell Culture–PCR**

INTEGRATED CELL CULTURE–PCR (ICC PCR)

- **Involves cell culture and PCR**
- **Biological amplification (cell culture) followed by enzymatic amplification (PCR)**

METHODS FOR DETECTION OF VIRUS

	Method of Detection		
	Cell culture	RT-PCR	ICC-PCR
Reduced time of detection	No	Yes	Yes
Infectious virus detected	Yes	Yes / No	Yes
Increased sensitivity	Yes	No	Yes
Affected by PCR inhibitory substances	No	Yes	No
Reduced costs	No	Yes	Yes
Detects only viable organisms	Yes	No	Yes
Detects viable but nonculturable virus	No	Yes	Yes

FACTORS AFFECTING TRANSPORT OF PATHOGENS THROUGH SOIL

- Size of microbe
  Transport of virus > bacteria > protozoa
- Soil texture
  Transport through sand > silt > clay
- Soil moisture—transport for saturated soils
  > unsaturated
- Change on microbes—generally negative
  ⑧ less sorption to negatively charged colloids
- Soil pH, generally greater than viral isoelectric point ⑧ viruses generally negative

TRANSPORT OF VIRUS IN BIOSOLID AMENDED SOIL

- Viral isoelectric point affects transport potential
- Most viruses negatively charged at neutral pH reducing sorption to colloids
- At lower (more acidic pH values) viruses are positively charged increasing sorption

FATE AND TRANSPORT OF PATHOGENS FROM BIOSOLIDS

- **Can pathogens from biosolids contaminate groundwater via transport through soil and vadose zone?**

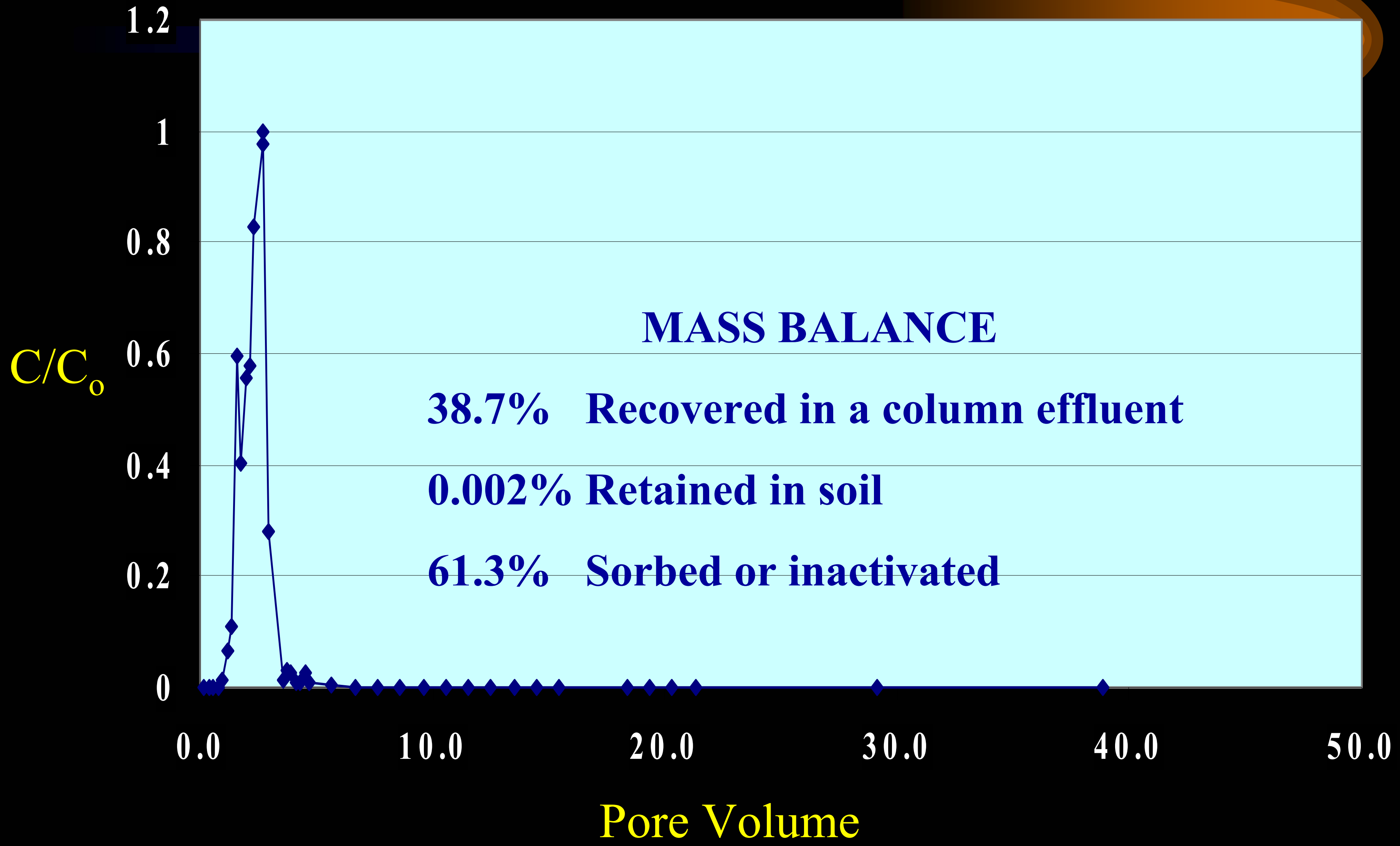
- **Does the presence of biosolids affect transport of virus?**

- **Most likely candidate for groundwater contamination: *virus***

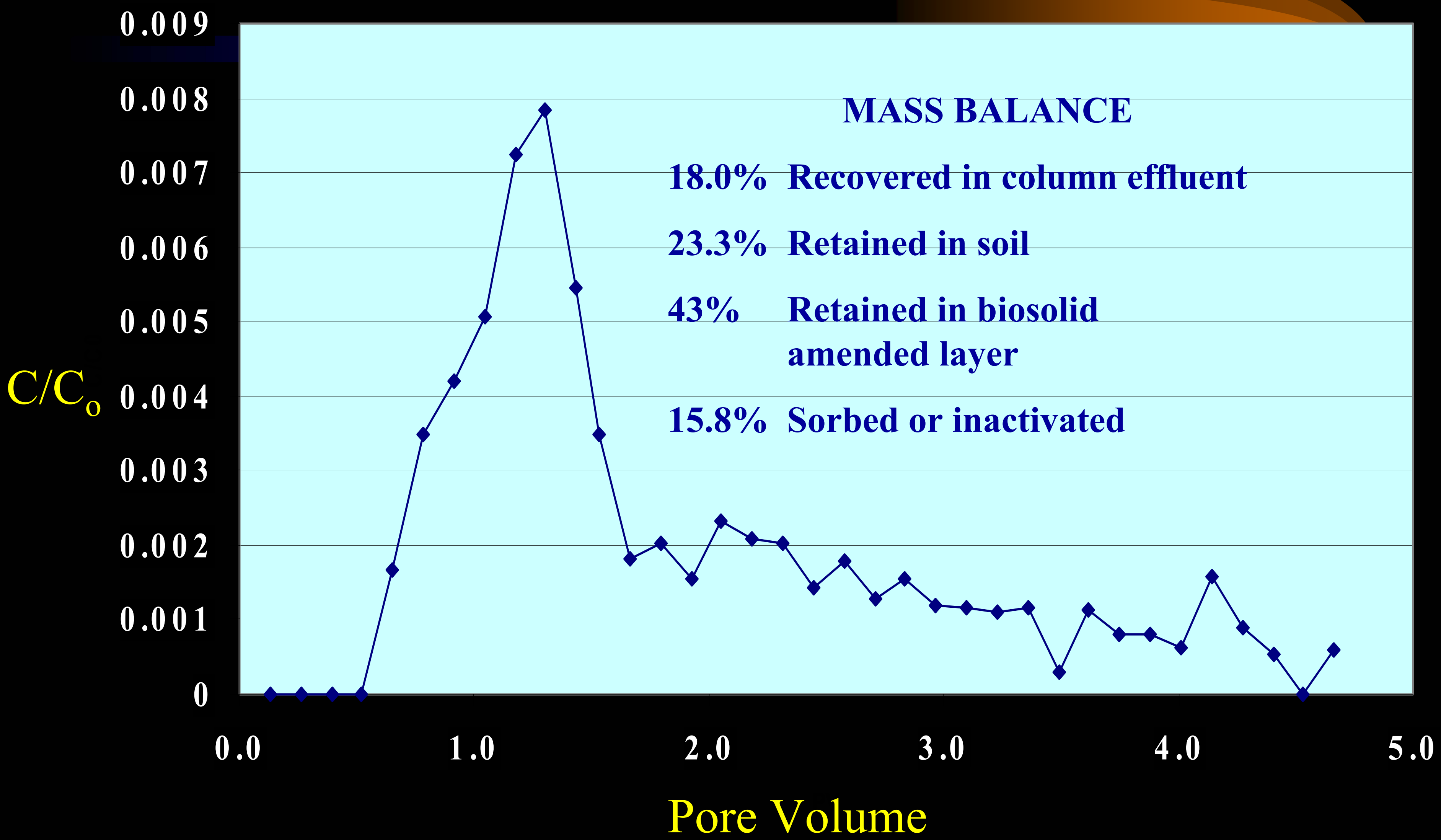
UNIVERSITY OF ARIZONA CURRENT TRANSPORT STUDIES

- Alexandra Chetochine—Studying for M.S. degree
- Column studies filled with Vinton sand
- Saturated flow
- Phage used as model for human enteric viruses
- Transport of pure cultures of phage studied, and phage from within biosolid

Experiment: MS2 Phage Transport When Seeded in Groundwater



Experiment: Biosolid Amended Soil MS2 Transport



FATE AND TRANSPORT—DISCUSSION

- Transport of phage from land applied biosolids less than from phage in pure culture
- Contamination of groundwater from phage unlikely
- U of A annual monitoring of groundwater from wells near land applied biosolids   negative (1985—Present)

FATE AND TRANSPORT OF PATHOGENS WITHIN LAND APPLIED BIOSOLIDS: SUMMARY

- If site restrictions and 503 regulations obeyed, little chance of direct contact with pathogens
- Two routes of exposure of concern with respect to pathogens, that can occur off site
  - bioaerosols
  - contamination of groundwater
- Both areas vigorously under study by The University of Arizona, National Science Foundation Water Quality Center