

Advances and Innovations to Achieve Microbially Safe and Sustainable Water

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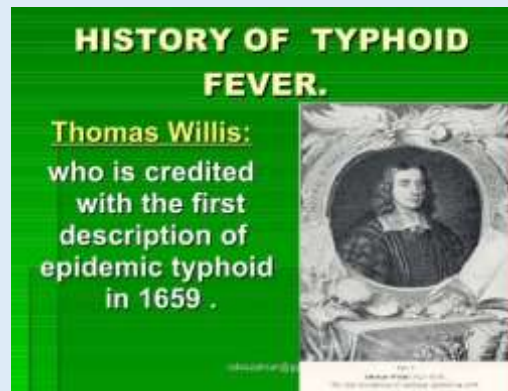
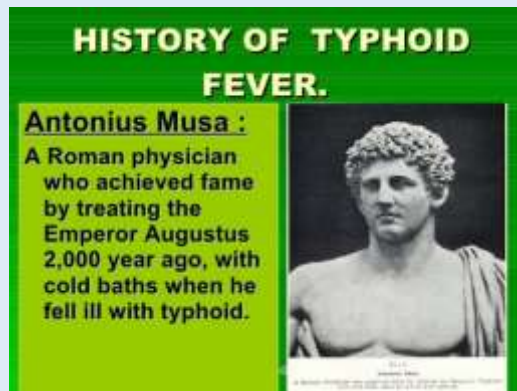
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Photos Courtesy World Health Organization-Geneva

Water, Sanitation and Infectious Diseases: A Brief History

- Adequate and safe water and effective sanitation were ancient concerns
- Cholera, Typhoid Fever, Dysentery, Hepatitis and Poliomyelitis were recognized diseases of old
- But these diseases were not attributed to water yet



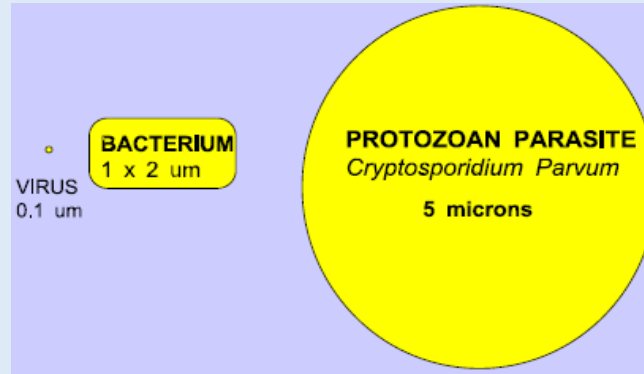
Polio paralysis



18th Dynasty (1403–1365 BC)
<https://commons.wikimedia.org/w/index.php?curid=1705155>

<http://www.slideshare.net/rabiezahran/typhoid-fever-3610355>

The Microbial World in Water: Breakthroughs of mid/late 1800s



- Microbes discovered using culture assays and other new tools
- Germ theory becomes documented
- Epidemiology links Infectious diseases to water, sanitation & hygiene
- Feces contaminating water identified as sources of bacteria causing infectious diseases
- Viruses as “filterable” microbes discovered (with ceramic microfilters)
- Enteric protozoan parasites identified

Water Quality Management Using Fecal Indicator Bacteria – Late 1800s

- *Escherichia coli* and related “coliform” bacteria found abundant in feces
- Fecal bacteria proposed as indicators of contaminated water
- Detect and quantify by culture methods
- Allowable limits for *E. coli* and coliforms set as zero/100 mL in USA and elsewhere
- Still used today



https://en.wikipedia.org/wiki/Agar_plate#/media/File:Escherichia_coli_colonies.png

Bacteria colonies on an agar plate



http://images.slideplayer.com/14/4503350/slides/slide_21.jpg

Multiple Fermentation Tube MPN Test

The Sanitary Engineering Revolution Helps Make Water Microbially Safe

Engineering principles were established by 1900:

- Choose water from a **safe source: no feces**
- **Treat water with:**
 - **chlorine** to kill bacteria
 - **filters** to remove bacteria
- **Protect** collected and treated water by **safe storage and delivery to user**

In principle, this system provides microbially safe water to consumers,sometimes

Question:

Despite the Sanitary Engineering Revolution of a century ago, why do so many people and communities still lack sustained access to safe water and sanitation and continue to suffer and die as a result?



My Answer:

Sustainable and consistent access to microbially safe water and sanitation for all requires:

- A better system - old one is not working
- New goals and targets
- A new framework
- New approaches and plans
- A broader, health risk-based focus
- New assessment and analysis tools
- A broader range of stakeholders

The 2015 Sustainable Development Goals

Provide new opportunities and approaches to achieve global access to safe water and sanitation for all by 2030



<http://steps-centre.org/engagement/beyond2015/>

Goal 6 for Water: Measure The Microbial Quality of Water to Track Access/Safety

- But, most water in the developing world is **never or only rarely tested** for microbial quality.
- If tested, it is **for wrong or uncertain reasons** that are **not actionable** at community and user levels.
- Microbial testing of water is **often inaccessible, too complicated and expensive.**
- ***A simple, self-contained, portable, lightweight, disposable and affordable test is needed.***

So, we created one!

Compartment Bag Test (CBT) for *E. coli* in water was developed to meet the need

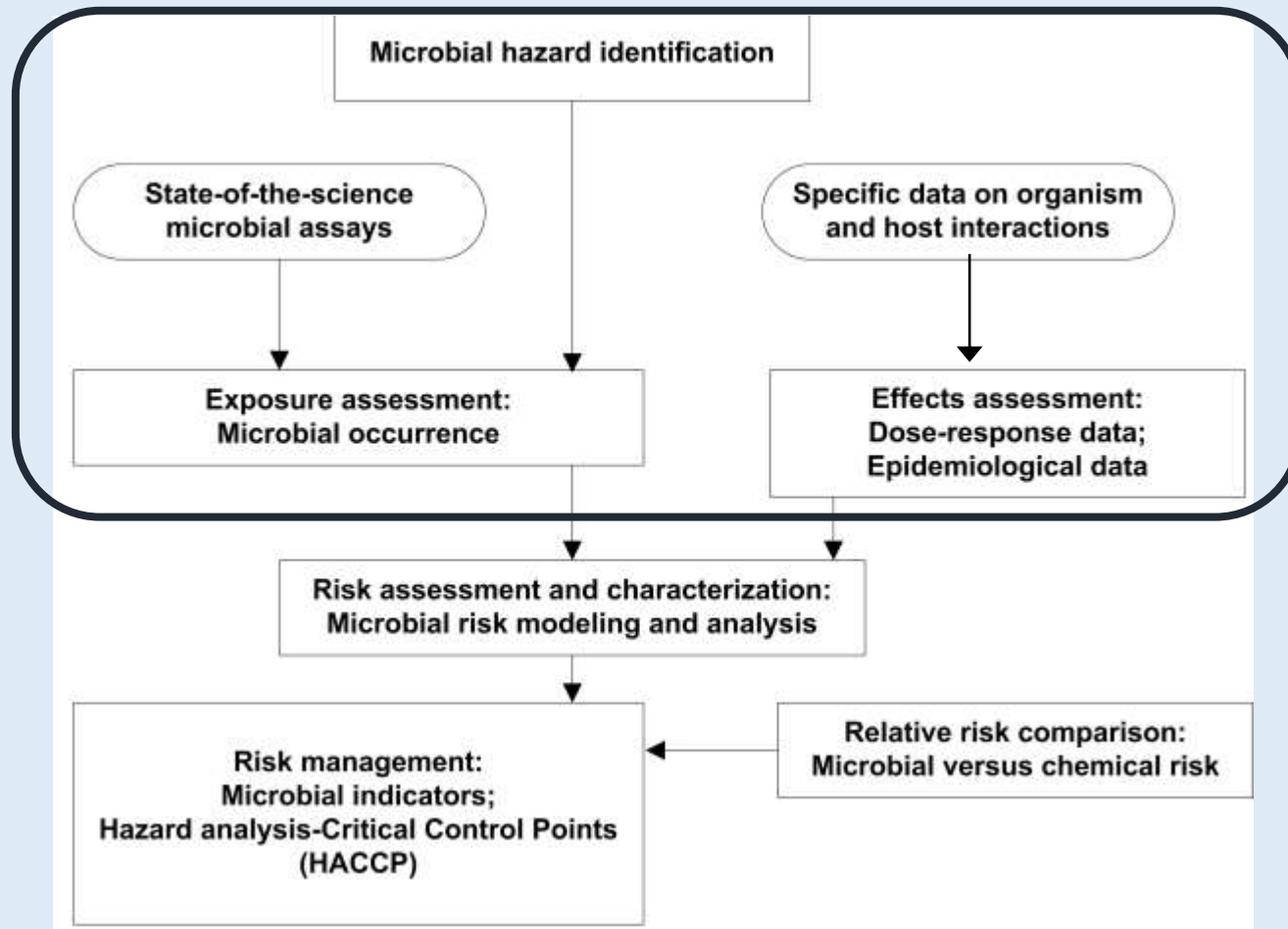
SIMPLE STEPS



Safety of Results	1 (10mL)	2 (30mL)	3 (56mL)	4 (3mL)	5 (1mL)	MPN/100mL	Upper 95% Confidence Limit Value/100mL
Unsafe	✓	✓	✓	✓	✓	>100	9435.1
Likely Unsafe	✓	✓	✓	✓	-	48.3	351.91
Possibly Unsafe	✓	✓	✓	-	-	13.6	83.06
Possibly Safe	-	✓	✓	-	-	4.7	22.75
Likely Safe	-	-	✓	-	-	1.5	7.81
Safe	-	-	-	-	-	0	2.87

Makes possible *E. coli* testing of water by anybody, anywhere, at any time, for any purpose

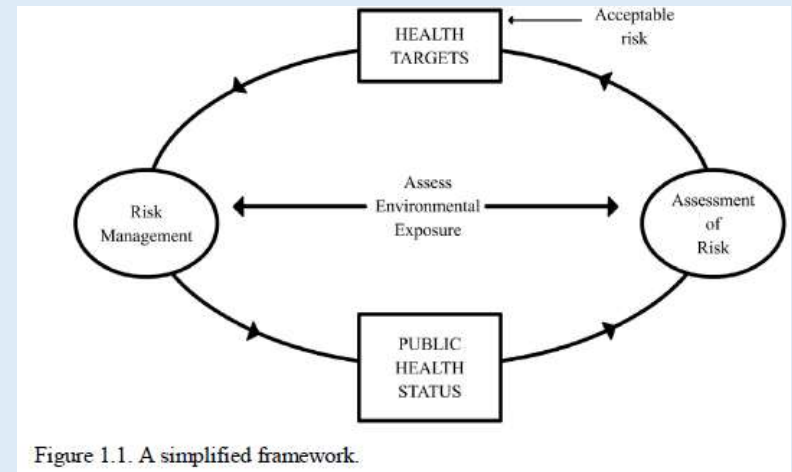
Quantitative Microbial Risk Assessment: Risk-based conceptual framework for assessing risks to health from microbes in drinking water



Sobsey, M.D., A.P. Dufour, C.P. Gerba, M.W. LeChevallier and P. Payment (1993) Using a conceptual framework for assessing risks to health from microbes in drinking water. *Jour. Amer. Waterworks Assoc.*, 85(3):44-48

The World Health Organization Stockholm Framework for Safe Water

- Holistic and actionable
- Multiple stakeholders
- Assesses public health status (surveillance)
- Assesses environmental exposure
- Incorporates risk assessment
- Sets health-based targets
- Integrates, informs and supports risk management
- Iterative

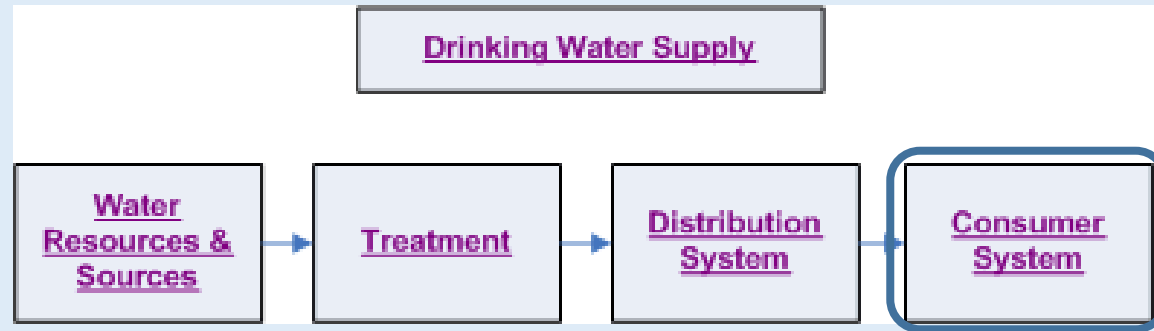


Water Safety Plans for Drinking Water

Overview

- Comprehensive risk assessment and management system
- Includes all steps from water source to consumer
- Based on principles and practices of the multiple barrier approach and HACCP (Hazard Analysis at Critical Control Points)

Drinking Water Supplies & Water Safety Plans



Water resources and sources

- Sources of raw water: surface, ground, harvested rain water, etc.

Treatment

- Chemical, biological or physical treatment of raw water

Distribution system

- Distribution via piping/other structures from storage to consumer

Consumer system

- Systems used to provide water to a household or other user beyond the point of delivery from the water supplier

What about people and communities without organized piped water systems or failed ones?

Household Water Treatment and Safe Storage (HWTS)

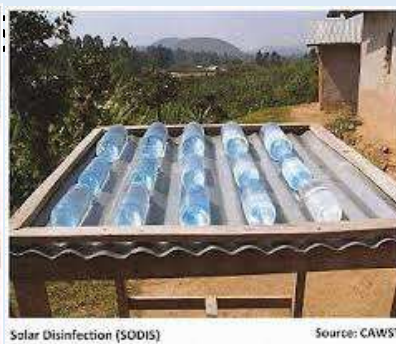
Treat and store water at point of use (POU) to make and keep it safe:



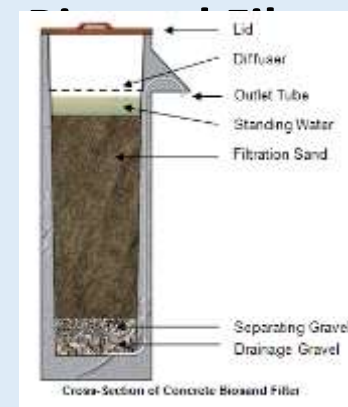
Source: ENPHO (2007)



Source: US CDC



Source: US CDC



<http://biosandfilters.info/technical/fact-sheet-biosand-filter>

Field Studies of Biosand Filters in Cambodian Households: Microbial Water Quality and Diarrheal Disease Evidence

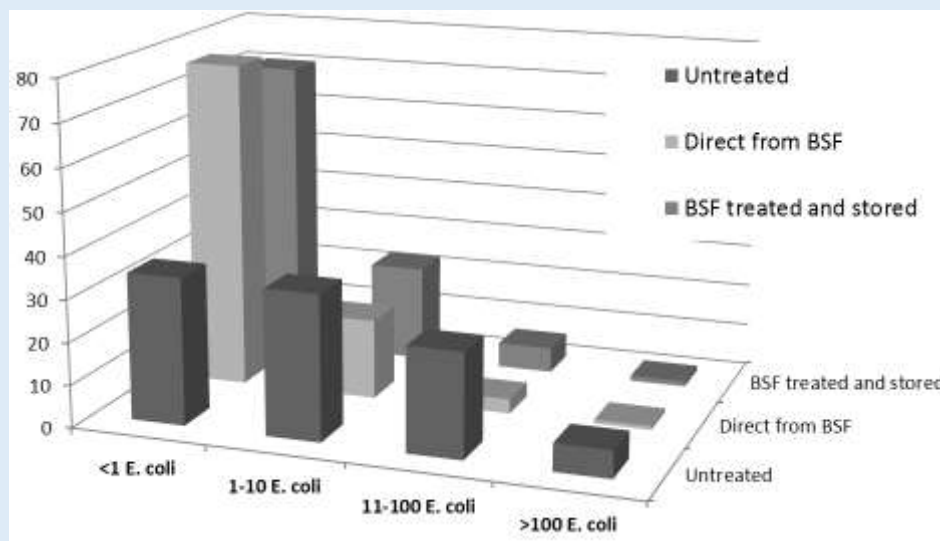
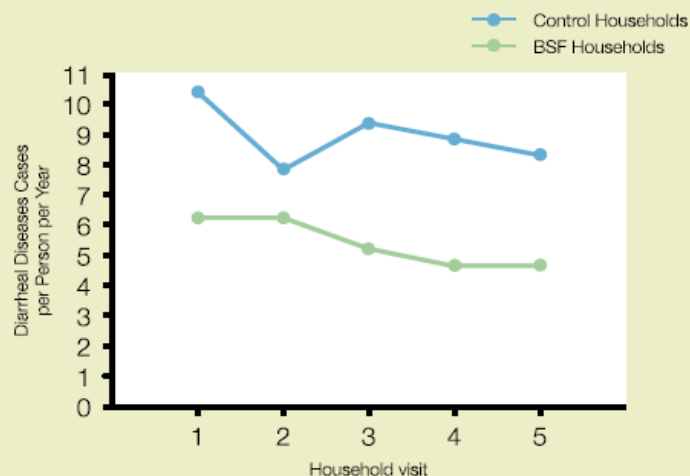


Figure 7: Cases of diarrhea per person per year by household group and visit (estimated from averages over five months)

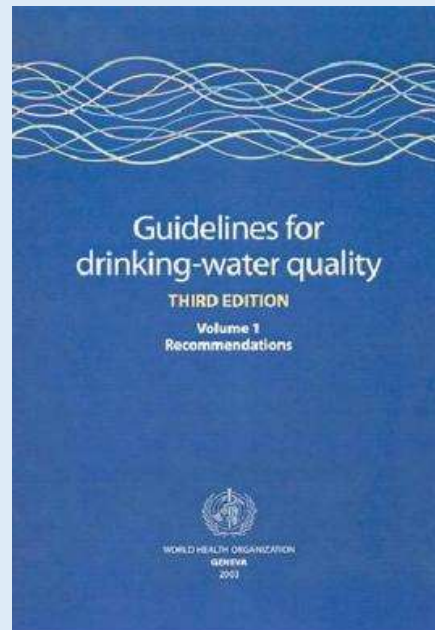
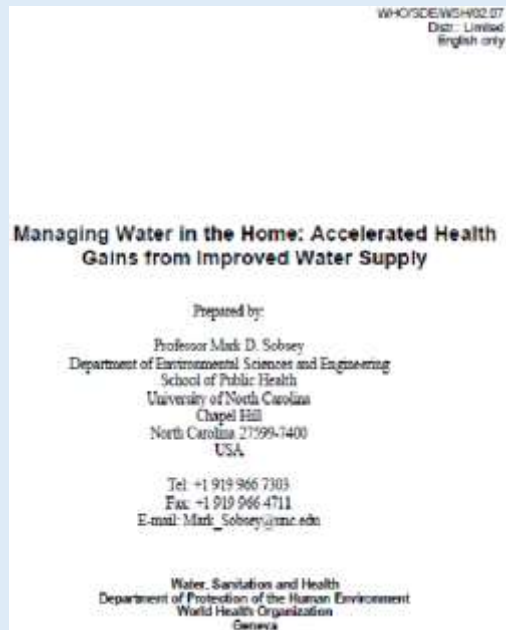


- Water quality as *E. coli*/100 mL was better in biosand filter (BSF) than in control households
- Diarrheal disease lower in BSF than in control households

Liang, K. et al., 2010) Use of BioSand Filters in Cambodia. Field Note, WSP, Cambodia.
https://www.wsp.org/sites/wsp.org/files/publications/WSP_biosand_cambodia.pdf

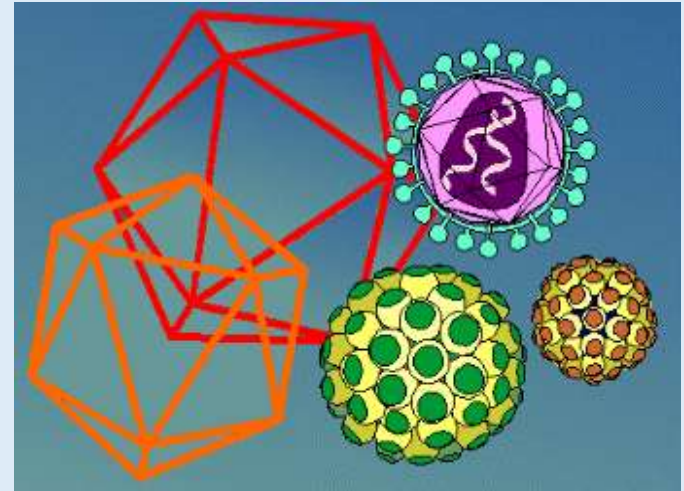
HWTS: Scientific Analysis Identifies Effective Technologies Leading to Guidance, Management & Use

- Reduces microbes in water
- Reduces risks of waterborne disease
- Recognized, supported and facilitated by the World Health Organization



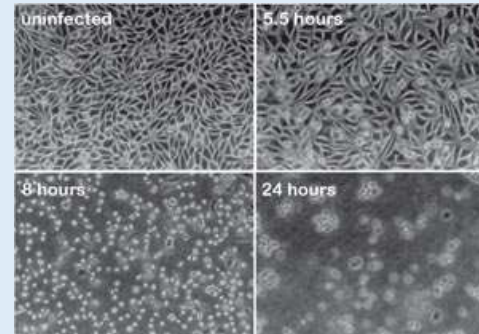
What about the Waterborne Enteric Viruses?

- Initial focus for waterborne diseases on bacteria: cholera, typhoid fever, dysentery
- Diseases from enteric viruses, like poliomyelitis, infectious hepatitis and viral gastroenteritis not addressed
- Culture of human enteric viruses in host cells became practical in the 1950s and 1960s
- Environmental virology emerged by 1965 - international conference: "Transmission of Viruses by the Water Route"
- My research began in 1966



Virus structure and composition

Poliovirus in host cells



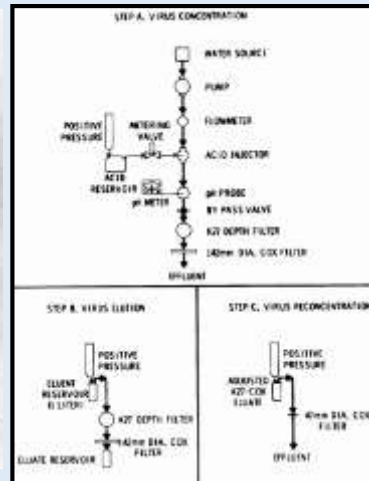
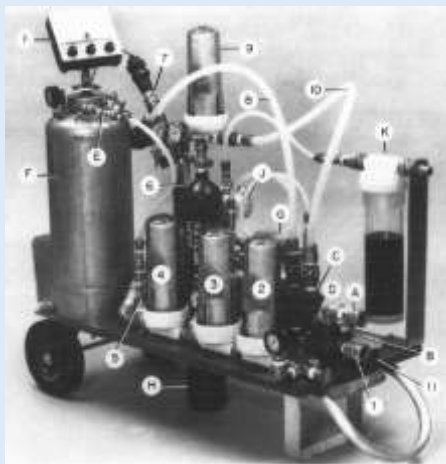
<http://www.virology.ws/2009/05/28/influenza-microneutralization-assay/>



A Need: Concentrate Viruses from Large Volumes of Water

1971-1974

- “Portable Virus Concentrator”
Baylor College of Medicine



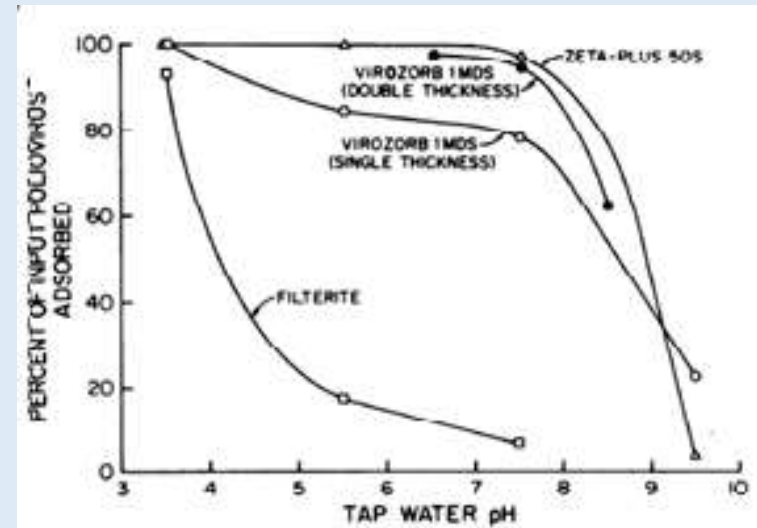
1996 to Present

- Hollow Fiber Ultrafilters
 - Hemodialysis filters repurposed
- Concentrates all pathogens in water



1975-1980 - UNC

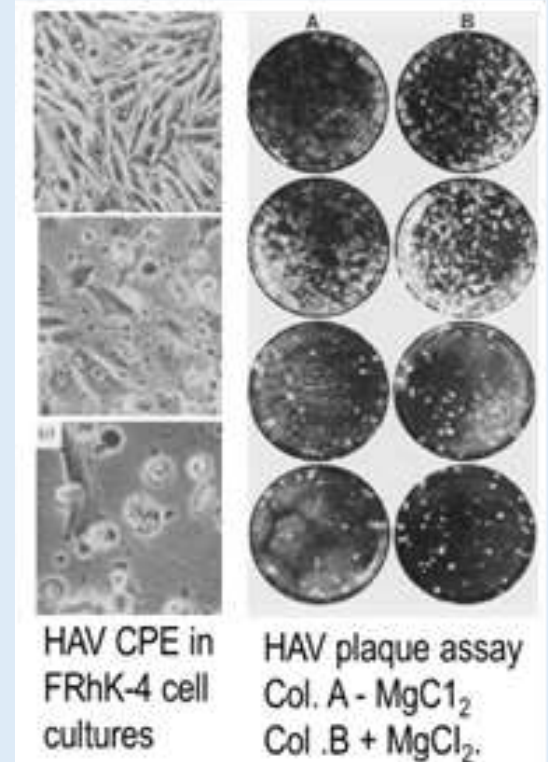
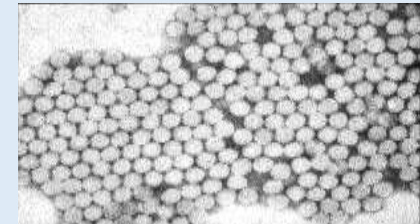
- Electropositive adsorbent filter



- Electropositive microporous adsorbent filter
 - Virosorb 1MDS
- Adopted by USEPA
- Used globally since ~1980

Addressing Hepatitis A Virus as a Waterborne Pathogen

- Identified and Isolated in 1973
- Grown in cell cultures in 1979
- Cytopathogenic strain isolated in 1987
- Simplified plaque assays for applied environmental studies

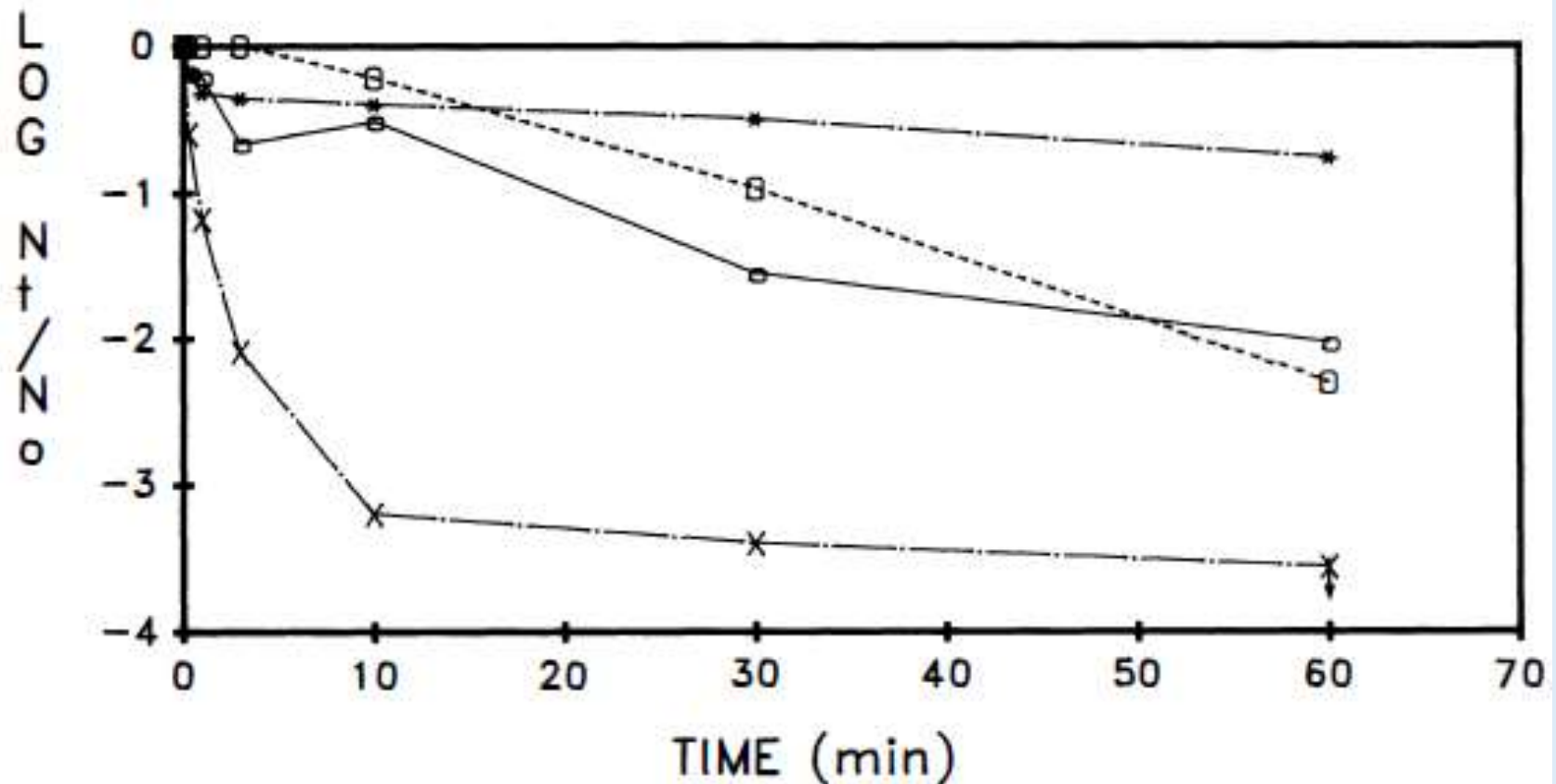


Cromeans, T., M.D. Sobsey and H.A. Fields (1987)
Development of a Plaque Assay for a Rapidly Replicating
Isolate of Hepatitis A Virus, J. Med. Virol., 22:45-56.

HAV Infectivity Assay in Cell Culture Makes Possible Key Environmental Studies

- Recovery and detection in water
- Waterborne outbreak investigation
 - HAV first recovered from water of an outbreak in 1981
- Survival in water & other environmental media:
 - Wastes, soils, sediments, strawberries & cookies(!)
- Reduction by disinfection and other treatment processes
 - for US EPA SWTR – CT values

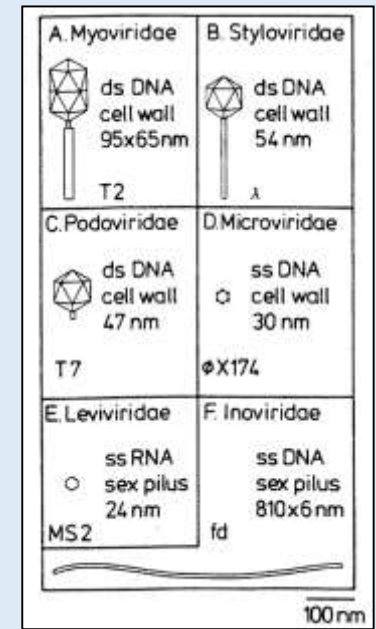
Figure 2. Inactivation of HAV (—□—), CB5 (---□---), MS2 (·—*—·) and OX174 (·—X—·) by 10 mg/l Monochloramine at pH 8 and 5°C. Arrow (↓) denotes the limit of virus detection.



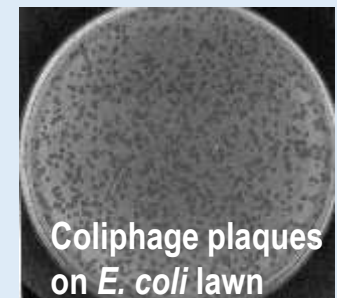
Sobsey, M.D., T. Fuji and P.A. Shields (1988) Inactivation of hepatitis A virus and model viruses in water by free chlorine and monochloramine, In: Proceedings of the IAWPRC Specialized Conference on Water and Wastewater Microbiology, Newport Beach, CA., Wat. Sci. Tech., 20(11 12):385 391.

Candidate Fecal Indicator Viruses to Address Risks from Waterborne Virus Pathogens: **Coliphages**

- Need a **fecal indicator virus for virus pathogens**
- **Coliphages**, viruses infecting *E. coli*:
 - Resemble human enteric viruses
 - Plentiful in feces and sewage
 - Survive and respond to treatment processes similar to human enteric viruses
 - Predict gastrointestinal illness risks from water
 - Easy & fast to detect and quantify like bacteria
- **Now regulated in groundwater**
- **Regulation in other waters is under consideration by US EPA**



IAWPRC (1991). *Water Res.* 25(5):529-545.



Coliphage plaques
on *E. coli* lawn

<http://www.sci.sdsu.edu/~smaloy/MicrobialGenetics/topics/phage/phage-lambda-plaque.gif>

Coliphages as Fecal Indicator Viruses

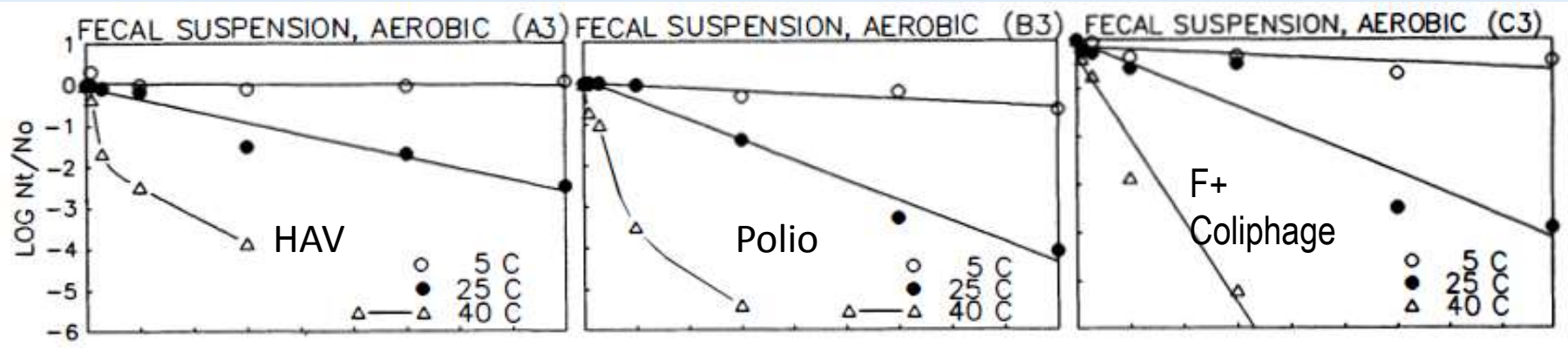
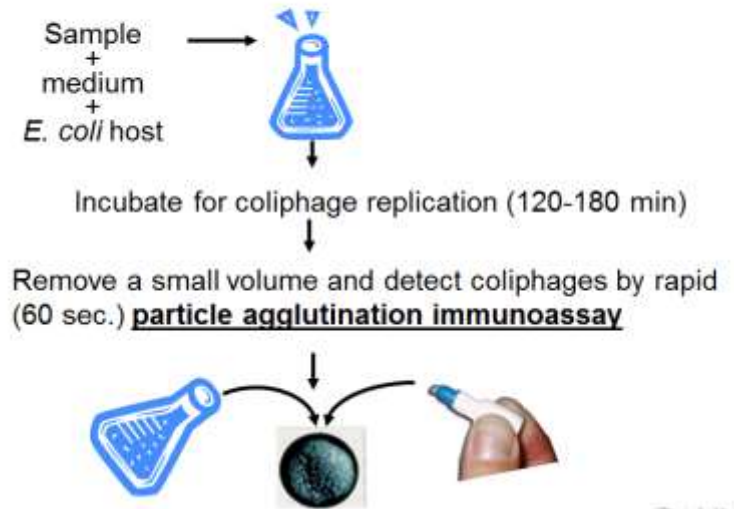
Potential for rapid detection:

- Broth enrichment culture
2-3 hrs
- Particle agglutination assay
1 minute

Love, D. C. and M. D. Sobsey. (2007) Simple and rapid F+ coliphage culture, latex agglutination, and typing assay to detect and source track fecal contamination. *Applied and Environmental Microbiology*. 73(13):4110-4118.

- Survive similar to virus pathogens

Rapid Coliphage Latex Agglutination Test (CLAT)



Gray, M., R. De Leon*, B. E. Tepper and M. D. Sobsey (1993) Survival of HAV, poliovirus and F-specific coliphages in disposable diapers and landfill leachates. *Wat. Sci. Technol.*, 27(3-4): 429-432.

The Future: We Need to Improve Technologies

- Piped water supplies and wastewater treatment systems are still lacking or deficient for too many.
 - How do we meet this dire need?
- Can **POU HWTS** be an effective, sustainable and accepted alternative where central piped water will never come?
 - Is it safe and sustained water access?
- What is sustained access to effective and safe **sanitation** and how do we achieve it?
 - Do we even know?



The Future: We Need to Do More on Social and Policy Aspects of WaSH

How to:

- **Overcome** entrenched and ineffective **bureaucracies** to improve policies, practices and regulations
- Identify and engage **other stakeholders with other skill sets** for a **team approach** to achieve the SDG
- Engage with and mobilize **consumers, users and others who must take responsibility for their water and sanitation**



My Microbiology “To Do” List Going Forward

- Direct **culture method for *Vibrio cholerae***, the cause of cholera
- Better culture methods to detect **indicators for viruses (coliphages) and protozoan parasites (*Clostridium perfringens*)**
- A “green”, non-toxic, biodegradable, natural organic polymer to **coagulate-flocculate** pathogens in water and wastewater: **chitosan**
- Encourage **microbial analysis of fecal wastes** to track & assess **SDG sanitation access**

My Microbiology “To Do” List Going Forward (continued)

- Detect and quantify key **antimicrobial resistant (AMR) bacteria** as “**AMR indicators**” in fecal wastes and contaminated waters
 - Occurrence, especially in “**hotspot**” **sources**
 - Human health risk assessment from environmental exposures
 - Assess impacts of interventions to reduce environmental release and presence

What is on Your “To Do” List Going Forward?

.....Thank-you!

Gratitude, Acknowledgements and Thanks

- To NWRI and the Joan Irvine Smith and Athalie R. Clarke Foundation
- To my mentors and teachers
- To my students, post-docs, visiting scholars and collaborators
- To my colleagues and friends
- To my family, most of all
 - Edie
 - My children Adam and Leah
 - My grandchildren, Lucas and Simon

