

# Human indicator persistence in the environment

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State-of-the-Science: Fecal Source Identification and  
Associated Risk Assessment Tools

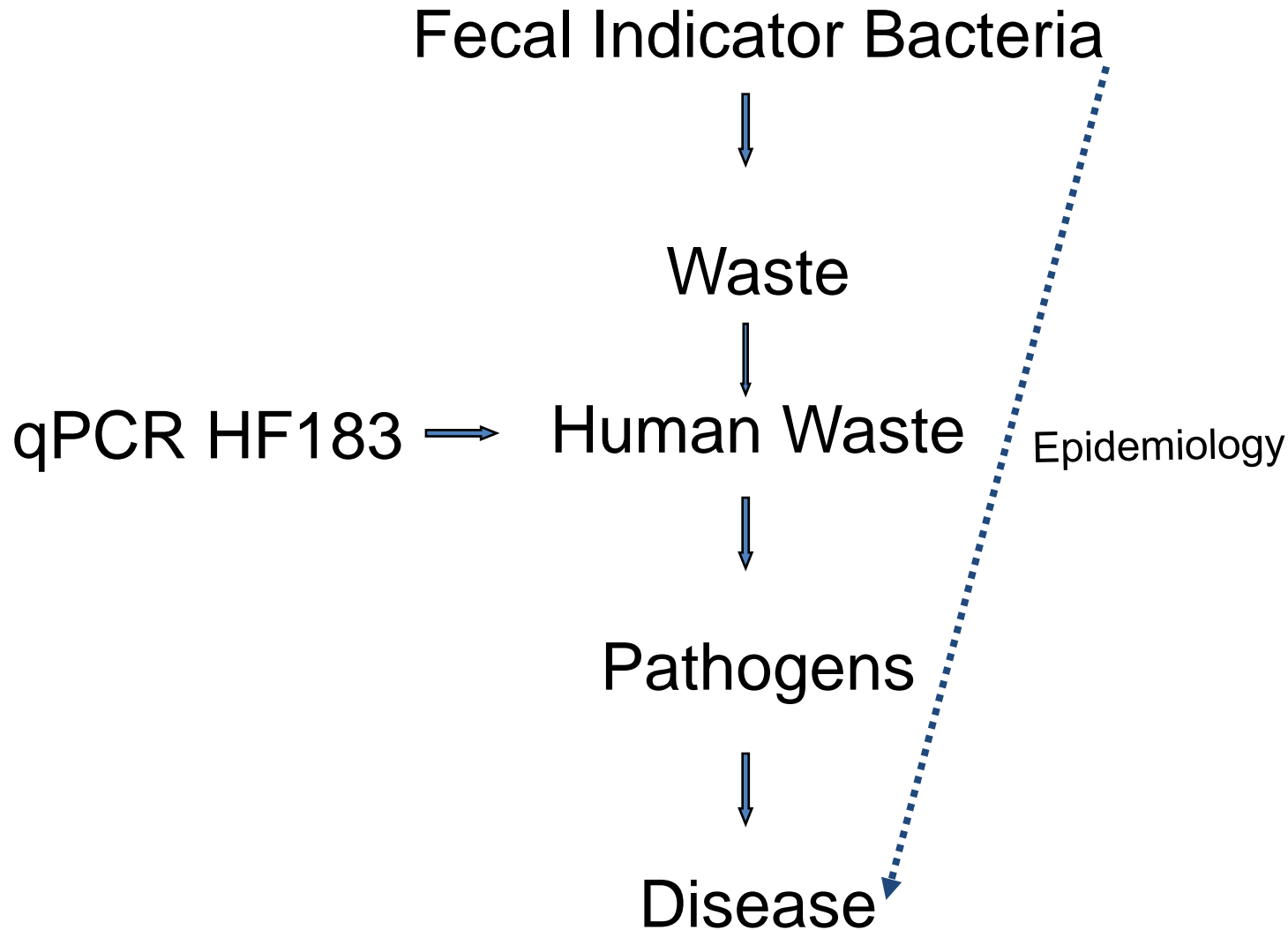
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Research Project Authority





# Fecal Indicator Bacteria: *a chain of inference*



# What is this HF183 “human marker”?

## DNA

- Extractable
- Amplifiable
- Specific to human waste

## Bacterial taxon

Domain: *Bacteria*

Phylum: *Bacteroidetes*

Class: *Bacteroidetes*

Order: *Bacteroidales*

Family: *Bacteroidaceae*

Genus: *Bacteroides*

Species: *dorei* (?)



(Haugland et al, 2010; Boehm et al., in rev. 2012)

(Carrero-Colón et al. 2010. Taxonomy, Phylogeny and Physiology of Fecal Indicator Bacteria. A chapter in **The Fecal Indicator Bacteria** edited by Michael Sadowsky and Richard Whitman. ASM Press. 328 pages; Haugland et al, 2010; Bakir et al. 2006 )

# HF183 Marker: Short History

- Discovery: Bernhard & Field (**2000**)
- X-method performance: Field et al. (**2003**)
- qPCR assay(s)
  - Seurinck et al. (**2005**)
  - Haugland et al. (**2010**)
- Comparison w/ other human markers, e.g.
  - Van De Werfhorst et al. (**2011**)
  - Boehm et al. (in rev. **2012**)

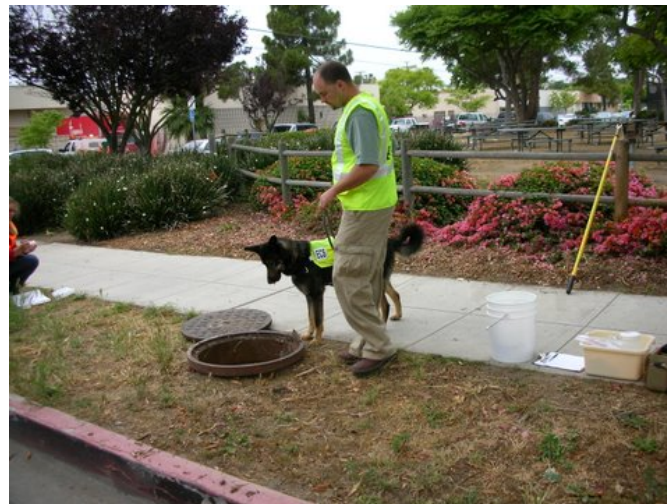
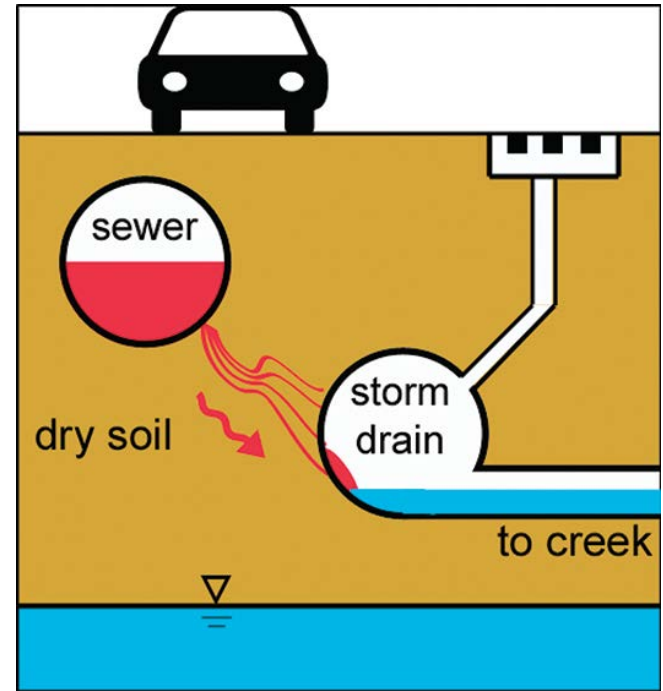
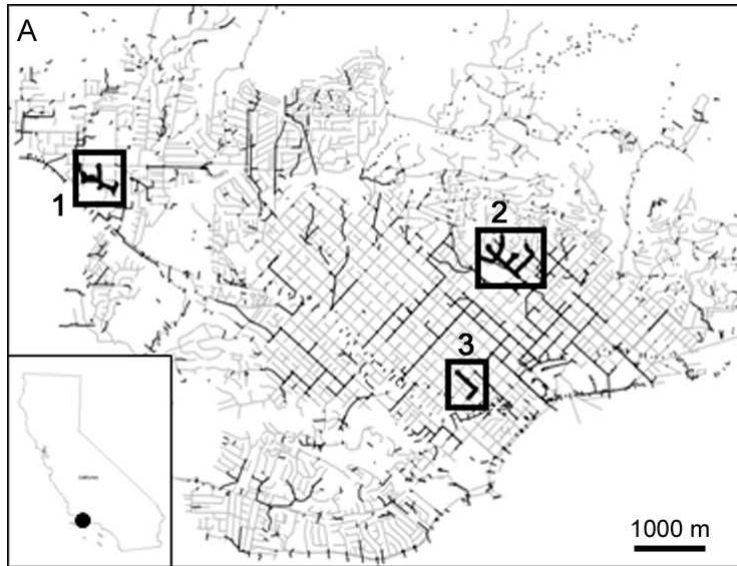
# Abundant in human fecal sources

- *Bacteroides* spp.
  - 30% of human fecal isolates
  - Human feces: up to  $10^{10}$  / gram (dry)
- HF183 marker
  - Sewage or septage:  $10^5$  to  $10^9$  copies / liter
  - Human feces:  $10^7$  copies / gram (wet)

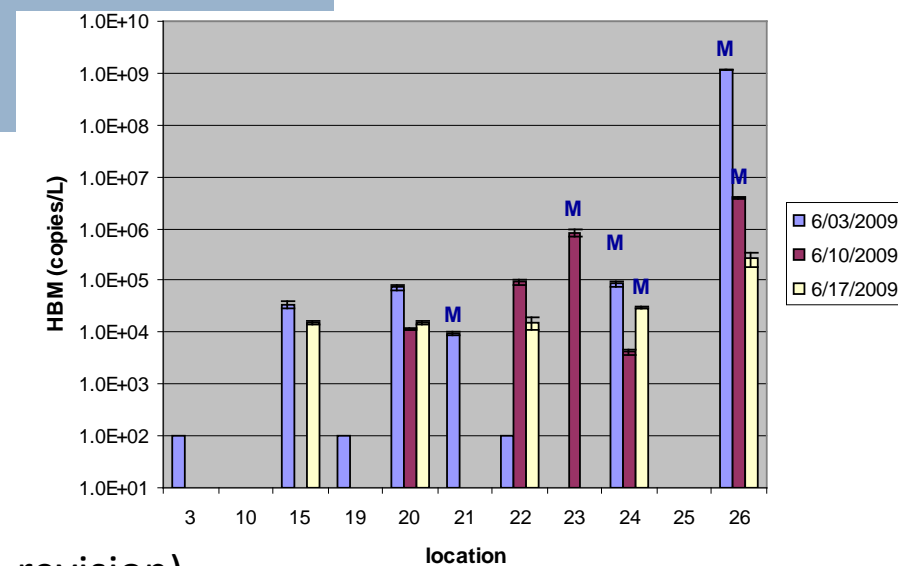
(Carrero-Colón et al. 2010. Taxonomy, Phylogeny and Physiology of Fecal Indicator Bacteria. A chapter in **The Fecal Indicator Bacteria** edited by Michael Sadowsky and Richard Whitman. ASM Press. 328 pagesHaugland et al., 2010; Layton et al., in rev. 2012)



# Application in Field Studies



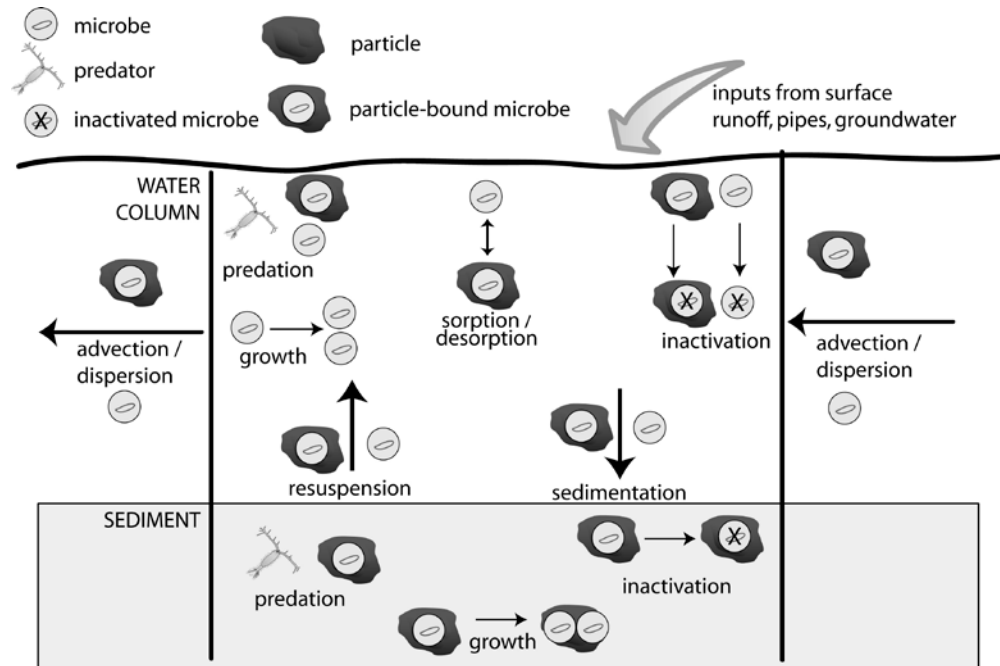
(Sercu et al., 2009; Sercu et al. 2011; Murray et al. 2011, WERF Report U2R09; Mission Study Report, 2011)





# Interpreting low levels of human marker in environmental waters

1. low levels are due to decay and/or dilution of target feces
2. low levels are due to cross reactivity with non-target feces (but requires lots of non-target feces in water if a



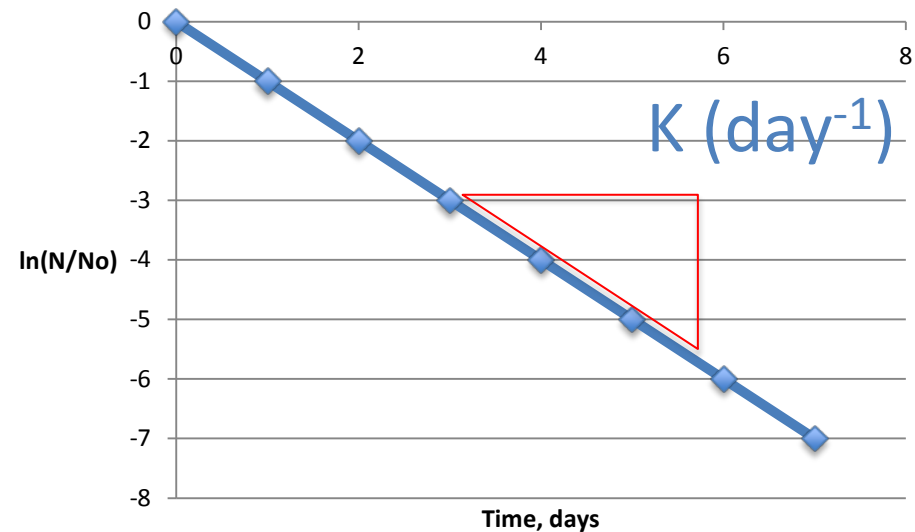
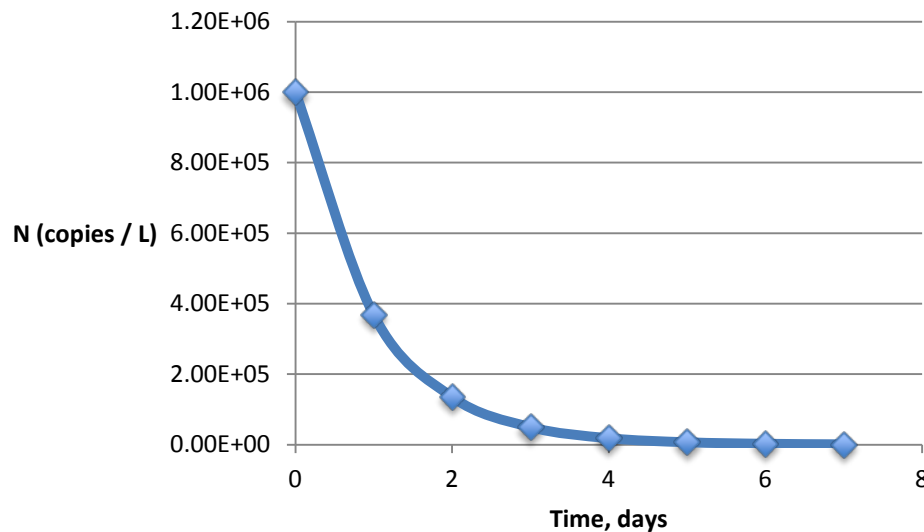
*We can't dismiss low levels of source-associated markers in environmental waters.*

(M. N. Nevers and A. B. Boehm, 2010. Modeling Fate and Transport of Fecal Bacteria in Surface Water.

A chapter in **The Fecal Indicator Bacteria** edited by Michael Sadowsky and Richard Whitman. ASM Press. 328 pages.)

# HF183: Rapid Decay

| Environment  | Location   | Reference             | K (day <sup>-1</sup> ) | Half life (d) |
|--------------|------------|-----------------------|------------------------|---------------|
| fresh-light  | Oregon-lab | Walters & Field, 2009 | -1.7                   | 0.4           |
| fresh-dark   | Oregon-lab |                       | -1.4                   | 0.5           |
| marine-dark  | Oregon-lab | Green et al., 2011    | -2.02                  | 0.3           |
| marine-light | Oregon-lab |                       | -0.25                  | 2.8           |
| fresh-dark   | Oregon-lab |                       | -1.34                  | 0.5           |
| fresh-light  | Oregon-lab |                       | -1.39                  | 0.5           |
| freshwater   | Florida    | Liang et al., 2012    | -0.73                  | 1             |
| seawater     | France     | Jeanneau et al., 2012 | -1.03                  | 0.7           |
| freshwater   | France     |                       | -1.39                  | 0.5           |



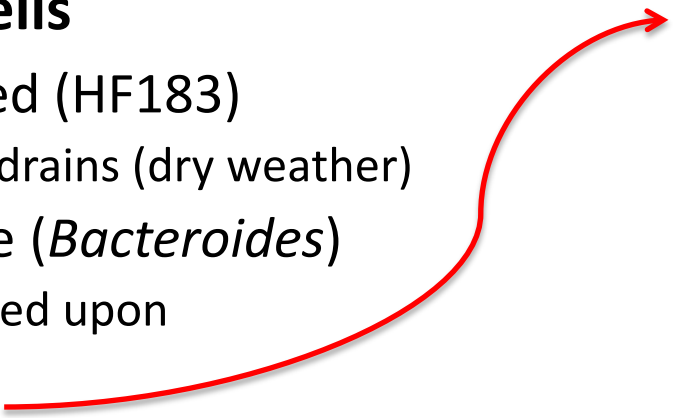
# Persistence scenarios

## Bacterial cells

- Introduced (HF183)
  - storm drains (dry weather)
- Attenuate (*Bacteroides*)
  - Predated upon
  - Die
  - Settle
- Establish
  - low diversity background (*E. coli*)
- Colonize (*Bacteroidetes* biofilms)
- Grow (*Bacteroides*)
  - nM oxygen (up to 5%)

## eDNA (generic)

- Accumulates
  - biofilms
  - soils
  - sediments
- Moves
  - even in soils, by capillarity
- Protected by clays
  - PCR-amplified from clays



(Van Elsas et al., 2012; Baughn et al. 2004; Meehan et al.; 2012; Gomez-Alvarez et al., 2012; Besemer et al.; 2009)

(Whitchurch et al. 2002; Steinberger et al. 2005; Pietramellara et al., 2009; Dell 'Anno et al., 2004; Ceecherini et al. 2007; Demaneche et al., 2001; Alvarez et al., 1998)

# Outstanding Issues

- Systematic understanding: decay in field conditions
- Relationship to regulated indicators, & pathogens
- How environmental factors govern
  - sunlight, temperature, salinity, predation,  $O_2$
- How to use data (field samples & conditions), plus knowledge of decay (rates, and factors) to diagnose (a condition or event)?
  - statistical approaches and/or modeling

# Moving forward

- SIPP MST studies: more field HF183 examples
- SIPP-proposed fecal “aging” study
  - Addressing open questions
  - Relating to pathogen taxa (community analysis)
- SIPP MST guide forthcoming
- However: HF183 marker is already useful in  
the context of a well-designed fecal source  
tracking study
  - Management actions need not wait



# Tools for Tracking Human Fecal Pollution in Urban Storm Drains, Creeks, and Beaches



- *Sources*
- *Tools*
- *Strategies*

*Dye entering Mission Creek (Santa Barbara, CA) after being flushed down toilets in nearby businesses. A private sewer lateral serving six businesses was found to be leaking into a storm drain.*

**City of Santa Barbara, Creeks Division**  
*In partnership with:*  
**University of California, Santa Barbara**

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(David Macaulay, 1976. *Underground*)