Risk-based thresholds for microbial source tracking markers

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There are a number of sensitive and specific fecal source-associated MST markers

- Human - HF183 Taqman, HumM2
- Ruminant - BacR, Rum2Bac
- Gull - LeeSeagull
- Swine - Pig2Bac




We have great tools for identifying host associated fecal bacteria

photo credit: wikipedia

Taking MST markers to the field…

Example result:

HF183 Taqman = BLOQ

[LOQ = 500 copies / 100 mL]

LeeSeagull = 3000 copies / 100 mL

enterococci = 100 CFU/100 mL

Cowell Beach, Santa Cruz, CA

What do these numbers mean?

We need guidance for allowable threshold concentrations of MST markers

Proposal: Risk-based thresholds

Is there enough human feces to represent a health risk?

Is there enough gull feces to represent a health risk?

1. *Is there enough human feces to represent a health risk?*

Research question

How does the concentration of human marker in recreational water relate to health risk if the source is raw sewage?

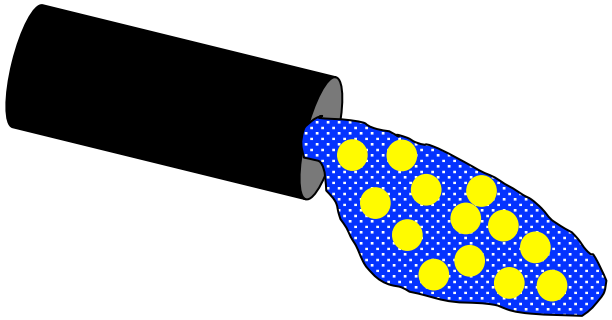
Approach: Use quantitative microbial risk assessment (QMRA)

QMRA scenario

- Raw sewage discharged into recreational waters
  - Raw sewage contains human markers and pathogens
- Swimmer is exposed to specific concentration of human markers
- Concentration of human markers is used to predict the amount of sewage in water
- Pathogens ingested with water while swimming
- Infection and illness risk predictions
  - dose-response
  - probability of illness given infection

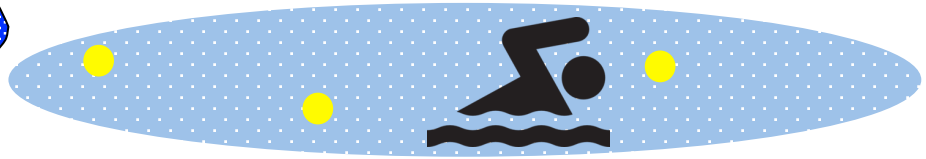
QMRA scenario

raw sewage with high density of human markers and pathogens



discharged into rec water

rec water with dilute sewage and human markers



swimmer exposed to human markers and pathogens

pathogen

water with human marker (white dot)

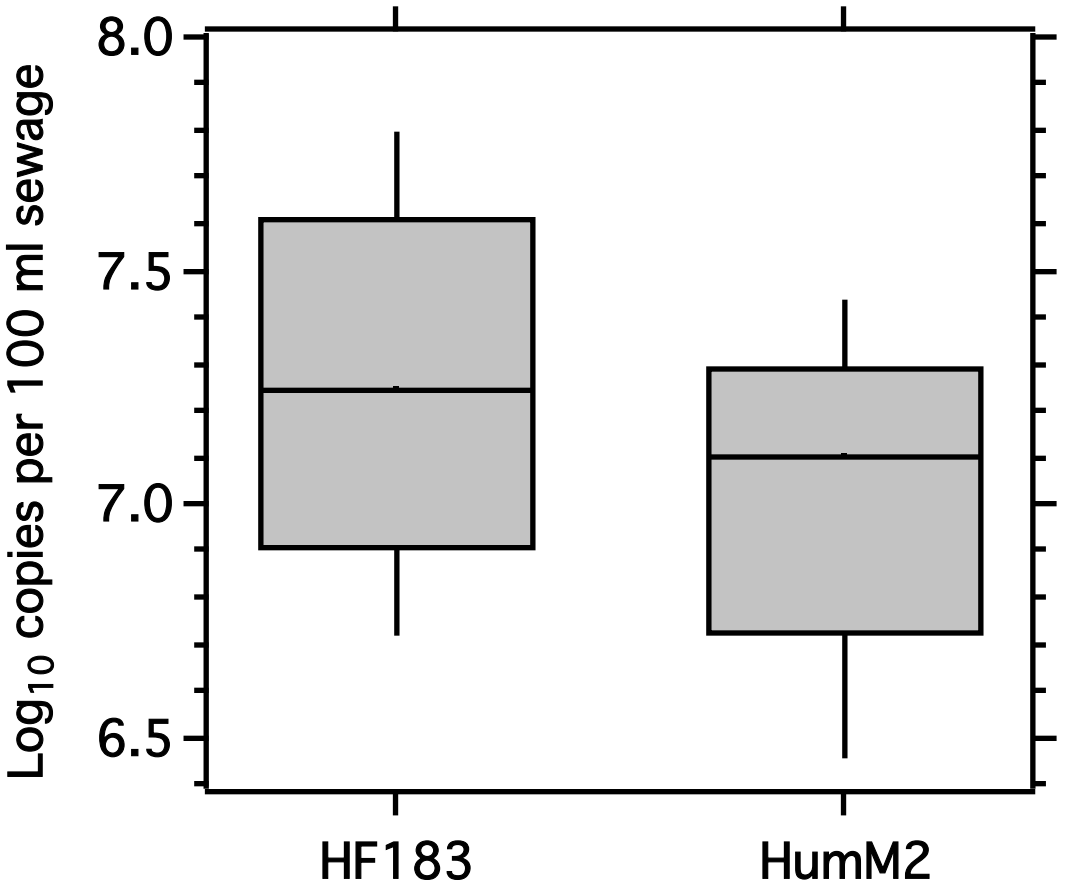
Example QMRA parameters

- Raw sewage has 10^7 copies / 100 ml human marker and 10^5 norovirus / 100 ml
- Human marker concentration is 10^3 copies / 100 ml at the beach
- Assuming human marker comes from raw sewage, concentration of norovirus is 10 norovirus /100 mL at the beach.
- Swimmer consumes 30 ml water
- Swimmer consumes ~ 3 norovirus
- Probability of infection is 0.4
- Probability of illness is 0.2

QMRA implementation

- Risk estimates for human marker at 1, 10, 100, 1000, 10000 copies/100 ml recreational water
- 10000 iterations per concentration using Monte Carlo simulations
- Model requirements:
  - volume of water ingested
  - human marker & pathogen concentrations in raw sewage
  - dose-response models and $P_{ill|infected}$
  - model parameters drawn from distributions
- Model output:
  - P_{ill_j} from each reference pathogen j
- $P_{ill} = 1\text{-}\Pi(1-P_{ill\_j})$

Distribution of HF183Taqman and HumM2 in raw sewage



54 samples of raw sewage from 37 states

Shanks et al. 2010. Environ. Sci. Technol. 2010, 44 (16) 6281– 6288

Reference pathogens in raw sewage

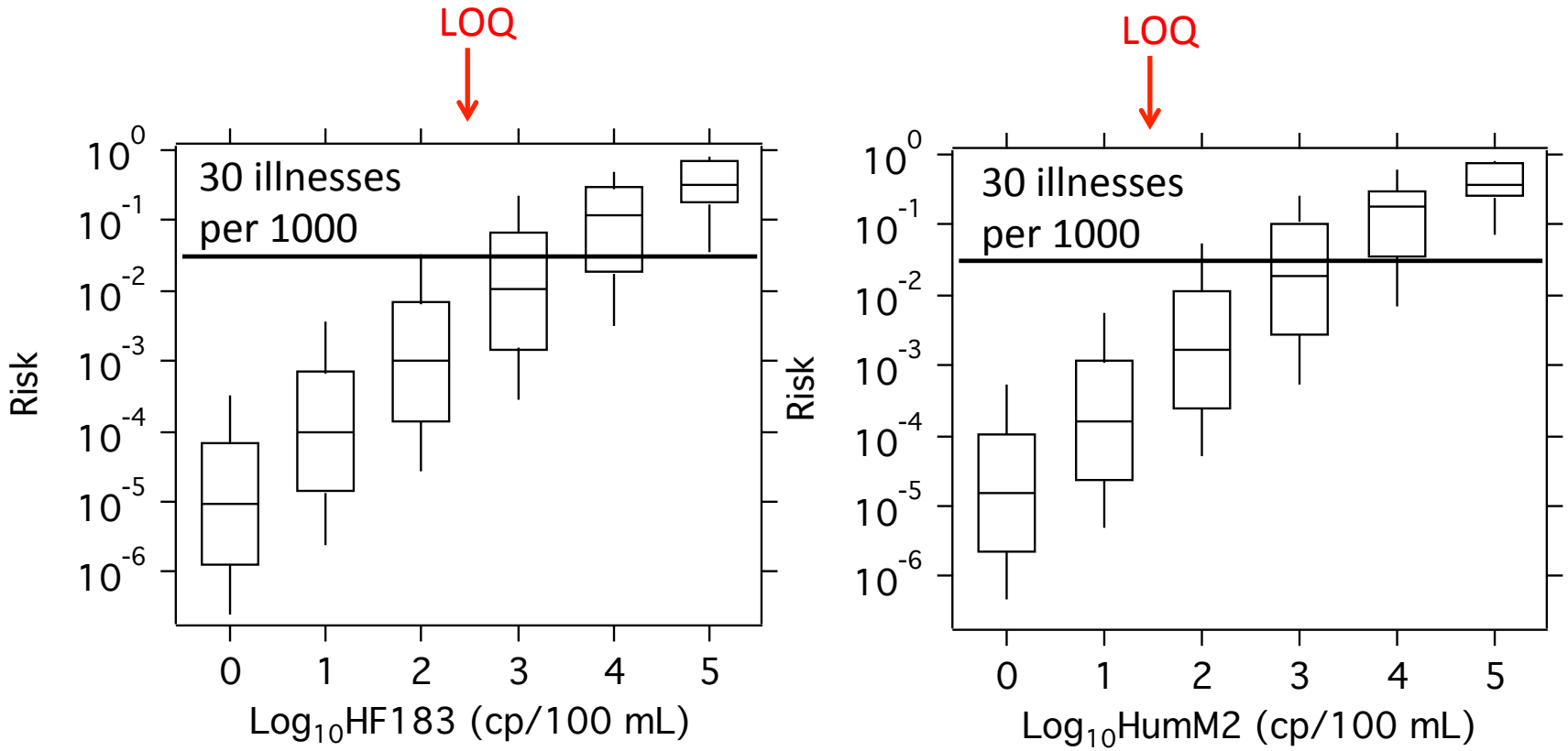
Organism	C_{sewage} range (log_{10} per L)	P_{inf}	$P_{ill|inf}$ (distribution)
Salmonella spp.	[0.5, 3]	$1-(1+ \mu/2884)^{-0.3126}$	0.17-0.4 (uniform)
Campylobacter	[2, 5]	$1- 1-{}_1F_1(0.024,0.024+0.011,-\mu)$	$1-(1+n\mu)^{-r}$
E. coli O157:H7	[-1, 3.3]	$1-(1+ \mu/48.8)^{-0.248}$	0.2-0.6 (uniform)
Cryptosporidium	[-0.3, 2.6]	$1 - \exp(-0.09\ \mu)$	0.3-0.7(uniform)
Giardia	[0.8, 4]	$1 - \exp(-0.0199\ \mu)$	0.2-0.7 (uniform)
Norovirus	[3, 6]	$1- {}_1F_1\ (0.04, 0.04+ 0.055, -\mu)$	0.6

Pathogens in raw sewage summarized in literature review by Soller et al. (2010) and Whiley et al. (2013), μ is dose

Volume ingested during swimming

log_e normal with mean of 2.92 and standard deviation of 1.43 units of ml (Dufour et al. 2006)

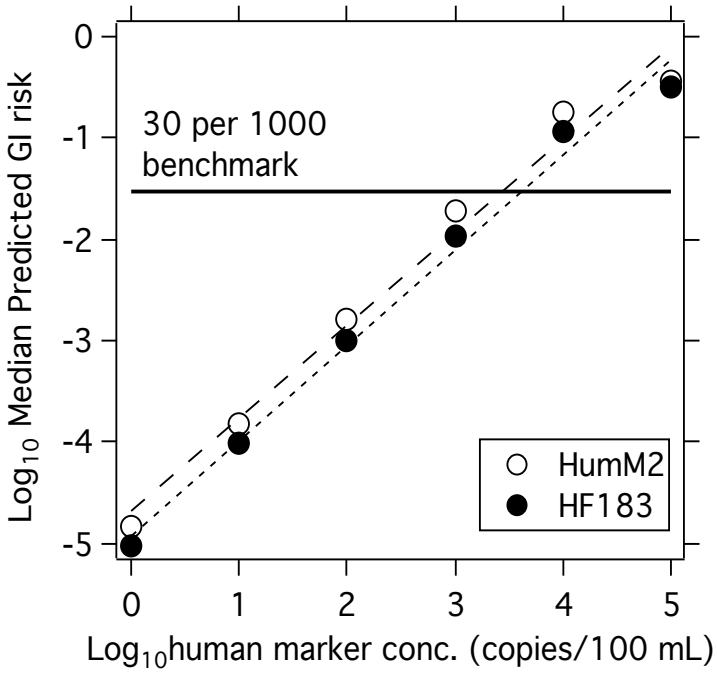
Results



Predicted health risk from all reference pathogens, but predicted health risk driven by norovirus

Three things to remember...

1. Human markers measured BLOQ in rec water could be risk relevant
2. 4200 copies HF183/100 ml gives rise to median GI risk of 30 per 1000
3. 2800 copies HumM2/100 ml gives rise to median GI risk of 30 per 1000



Boehm et al. 2015. *Environmental Science & Technology Letters*, 2, 270-275.

What if the source of human marker is treated effluent?

Treated effluent has a different concentration profile of pathogens and human markers than raw sewage

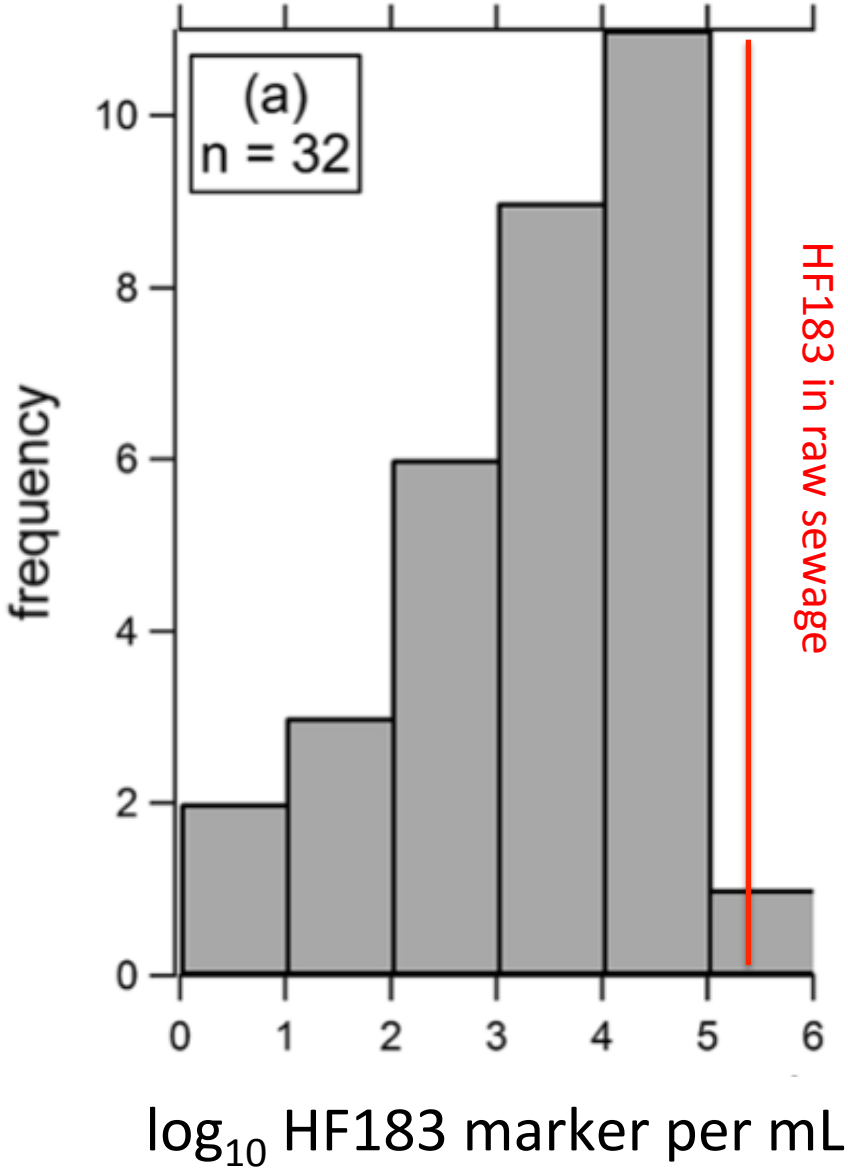
Research question

How does the concentration of human marker in recreational water relate to health risk if the source is treated effluent?

Use same QMRA approach

HF183 marker in treated effluent

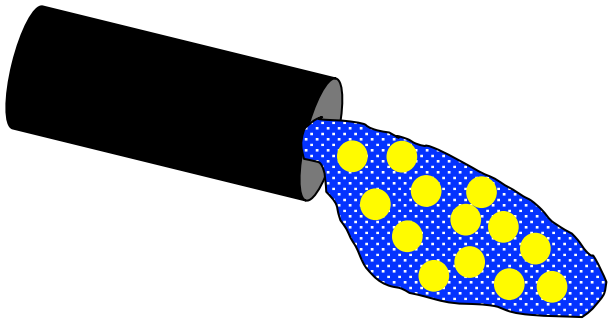
- Visited 32 wastewater treatment plants in California
- Final effluent collected at each plant
- HF183 marker measured using QPCR



Pathogens in treated effluent

Organism	C_{sewage} range (log_{10} per L)	$C_{effluent}$ range (log_{10} per L)
Salmonella spp.	[0.5, 3]	ND
Campylobacter	[2, 5]	ND
E. coli O157:H7	[-1, 3.3]	ND
Cryptosporidium	[-0.3, 2.6]	[-1.3, 1.6]
Giardia	[0.8, 4]	[-1.3,2.8]
Norovirus	[3, 6]	LR= -4.6, -1.1

treated effluent with human markers and pathogens



discharged into rec water

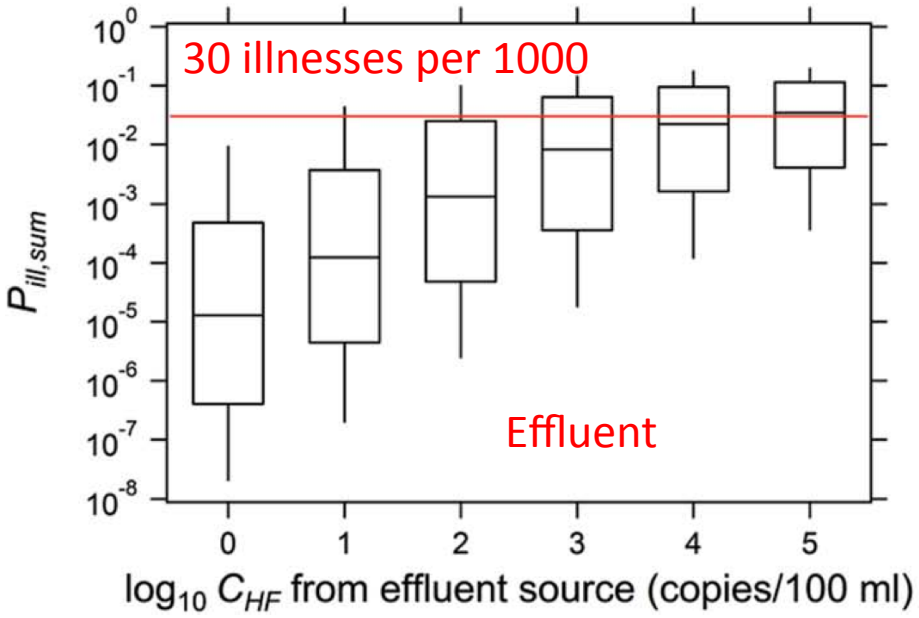
rec water with dilute wastewater and human markers

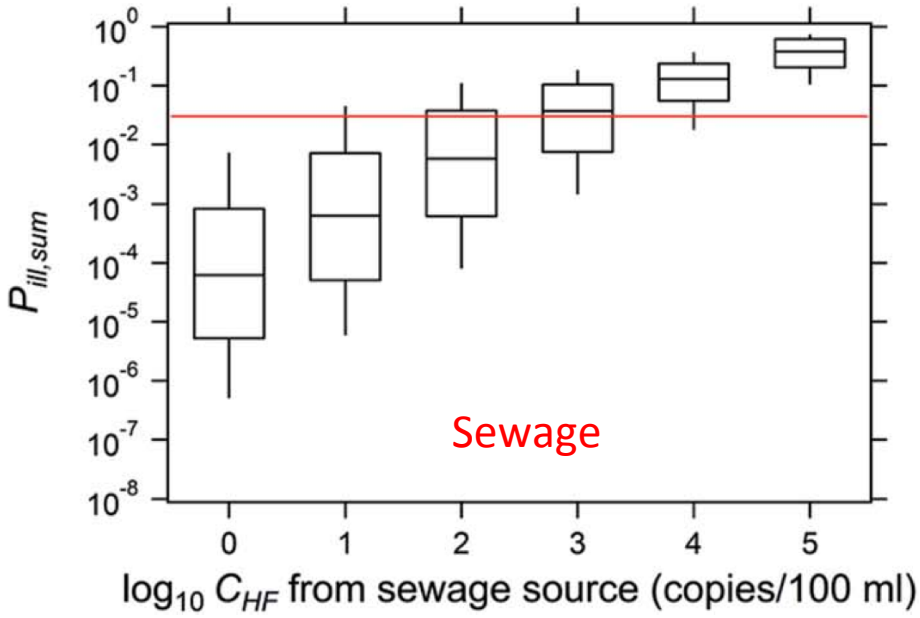
swimmer exposed to human markers and pathogens

pathogen

water with human marker (white dot)

Risk as a function of HF183 concentration





Two things to remember….

1. There is HF183 in treated effluent
2. 20,000 copies HF183/100 ml gives rise to median GI risk of 30 per 1000 if source is treated effluent

Brown et al. 2017. *Environmental Science: Processes and Impacts*, In press.

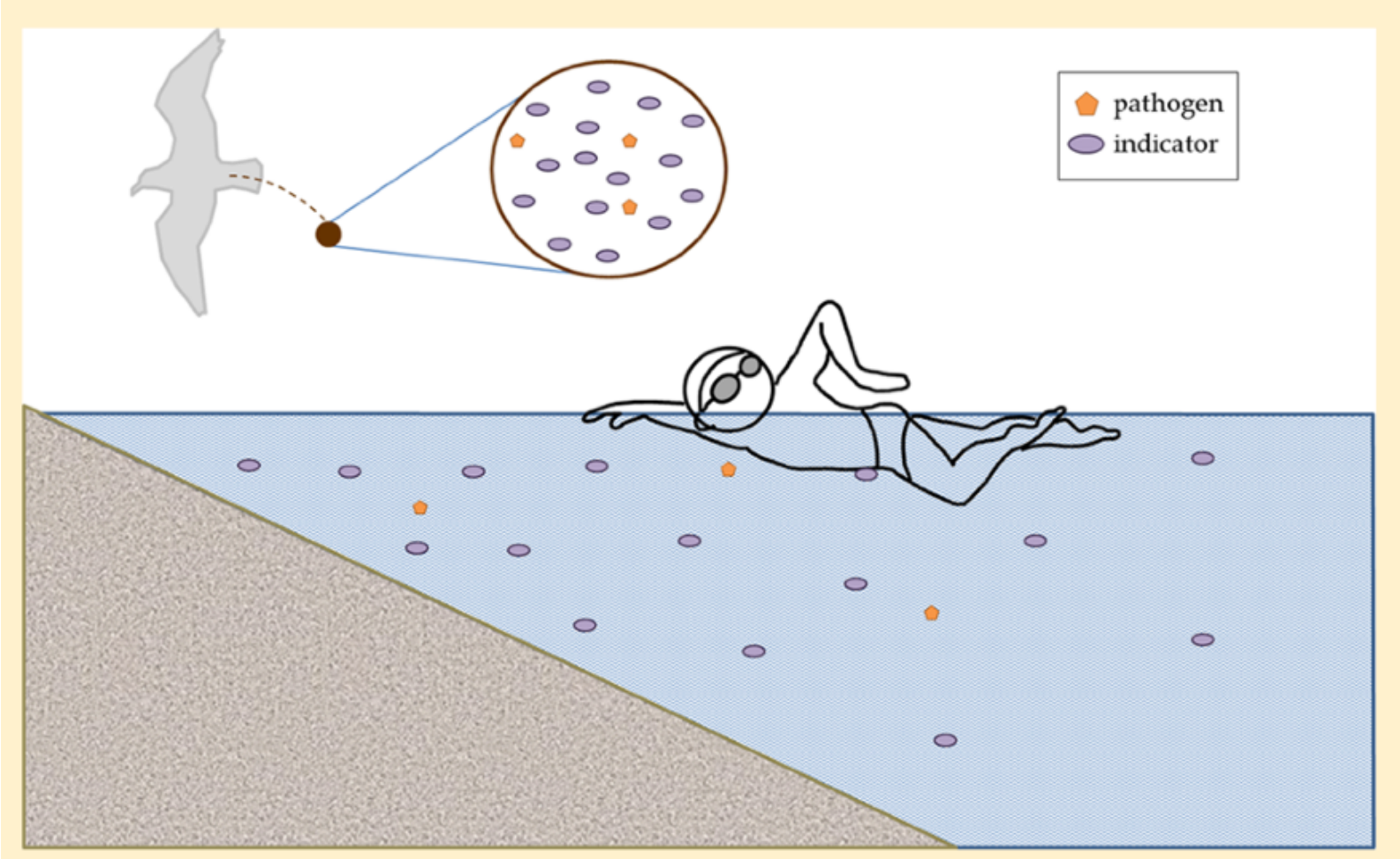
2. *Is there enough gull feces to represent a health risk?*

Research question

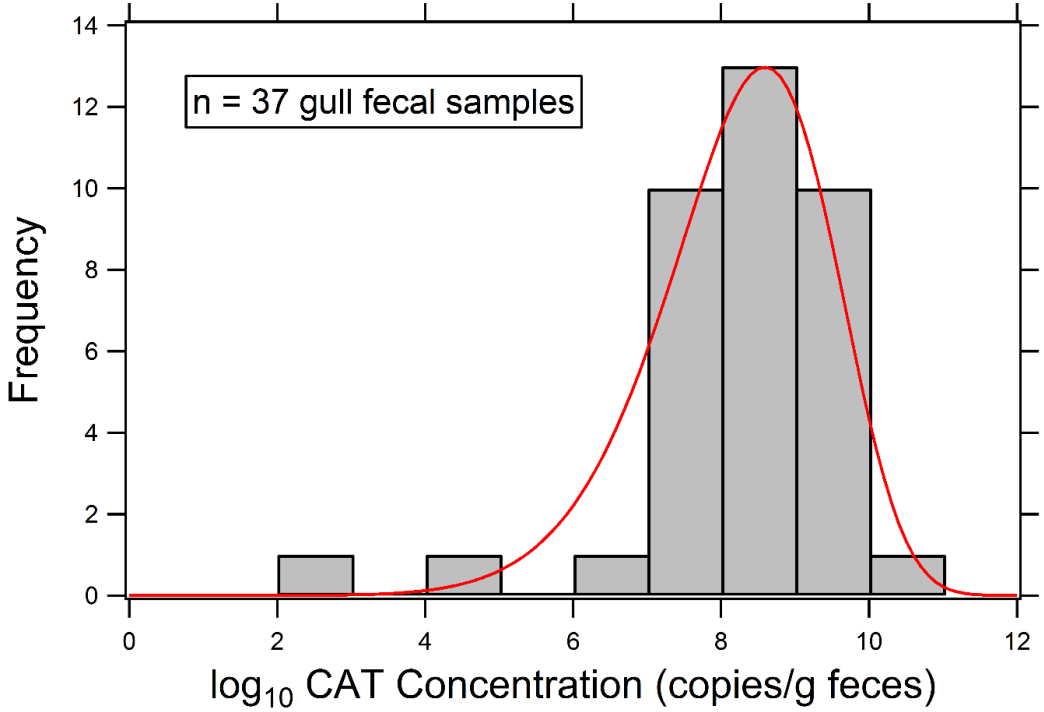
How does the concentration of gull marker in recreational water relate to health risk?

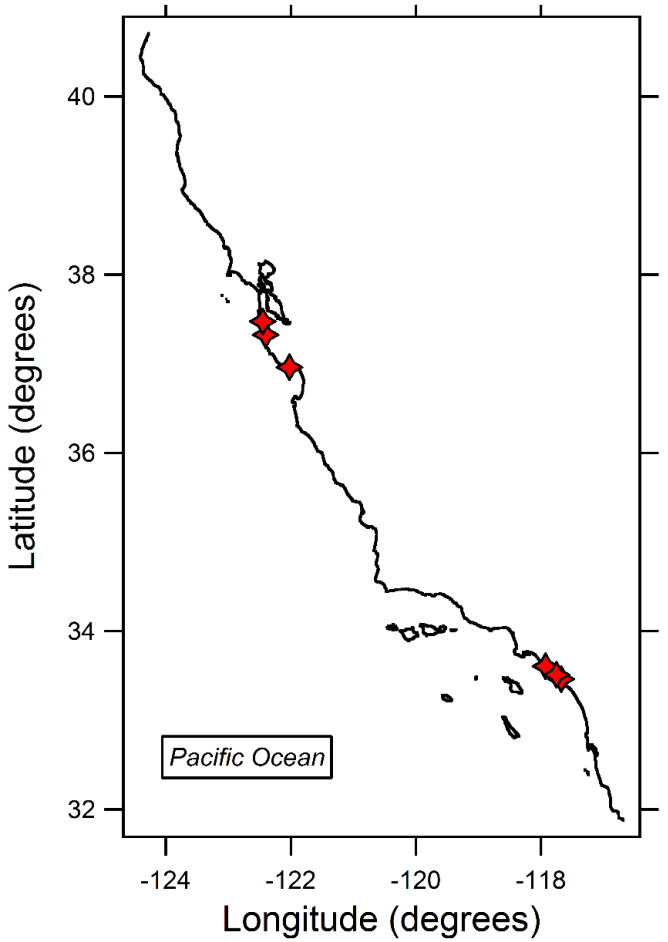
Use same QMRA approach

Scenario



Gull marker (CAT) in California gull feces

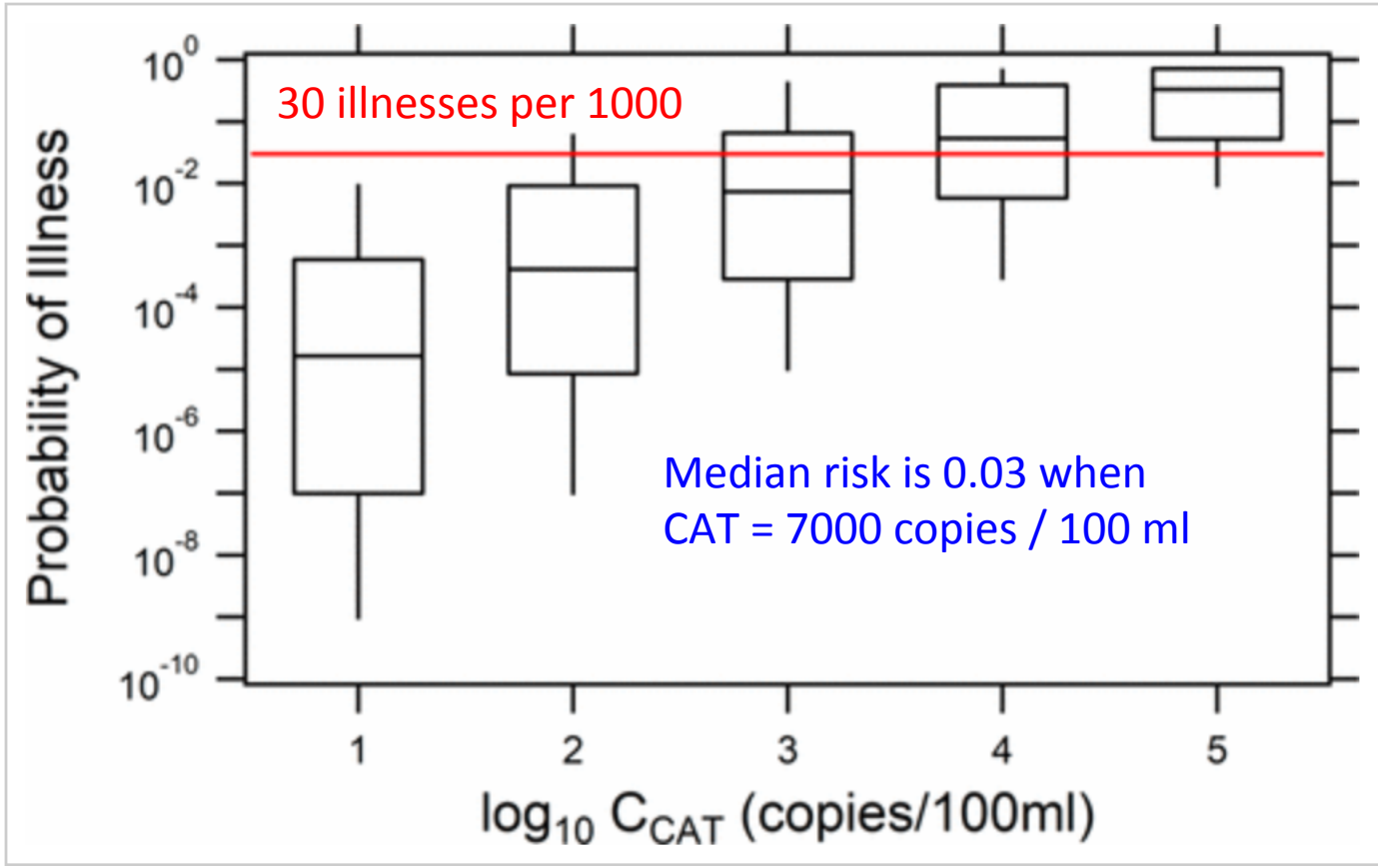




Pathogens in gull feces

Organism	C_{sewage} range (log_{10} per L)	$C_{effluent}$ range (log_{10} per L)	C_{gull} range (log_{10} per g)
Salmonella spp.	[0.5, 3]	ND	[2.3, 9.0]
Campylobacter	[2, 5]	ND	[3.3, 6.0]
E. coli O157:H7	[-1, 3.3]	ND	ND
Cryptosporidium	[-0.3, 2.6]	[-1.3, 1.6]	ND
Giardia	[0.8, 4]	[-1.3,2.8]	ND
Norovirus	[3, 6]	LR= -4.6, -1.1	ND

Risk as a function of CAT concentration



This result changes dramatically if you use a different Campylobacter *dose-response function, by 2 orders of magnitude*

Two things to remember….

1. Choice *of Campylobacter* dose-response function can make a big difference.
2. Using the "dose-dependent" relationships, risk based threshold for CAT is 7000 copies / 100 ml.

Brown et al. 2017. *ES&T Letters*.

What if we have a mixture of two different sources?

<u>Example result:</u>

HF183 Taqman = BLOQ

[LOQ = 500 copies / 100 mL]

LeeSeagull = 3000 copies / 100 mL

enterococci = 100 CFU/100 mL

Cowell Beach, Santa Cruz, CA

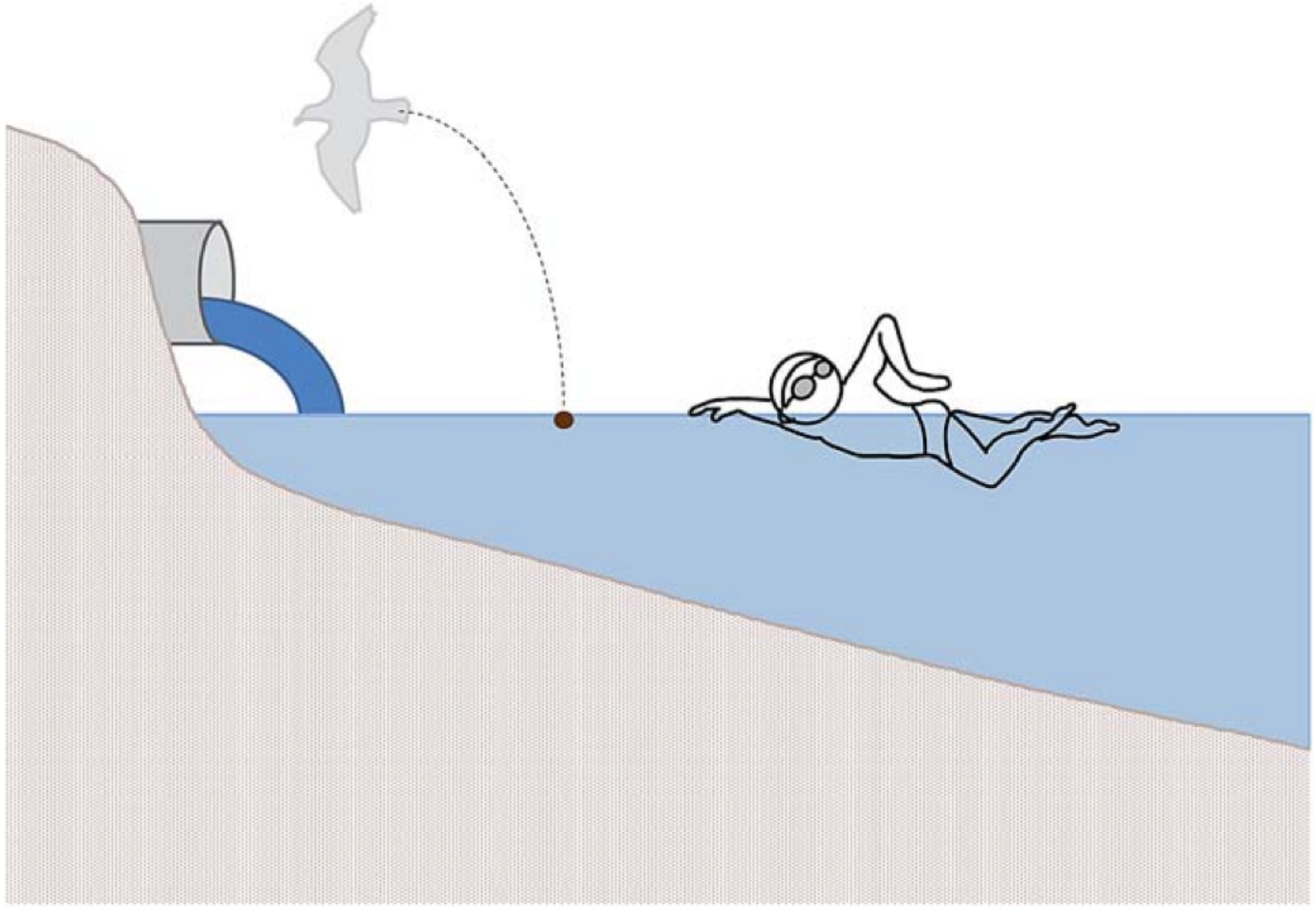
QMRA scenario can consider both sources additively in calculating a dose

Research question

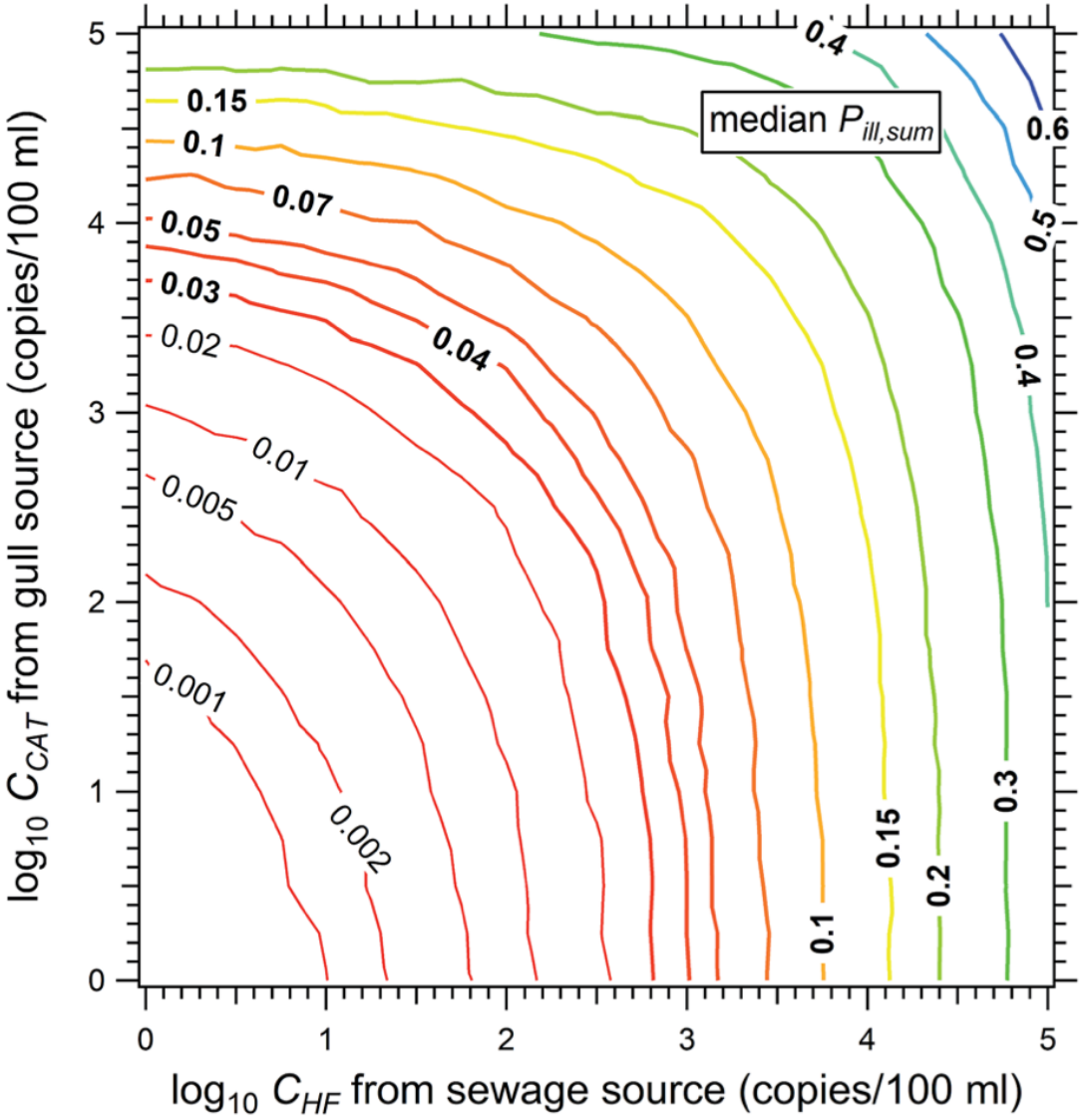
How do two sources (feces from gulls and raw sewage) of fecal pollution to recreational waters interact to affect risk?

QMRA scenario can consider both sources additively in calculating a dose

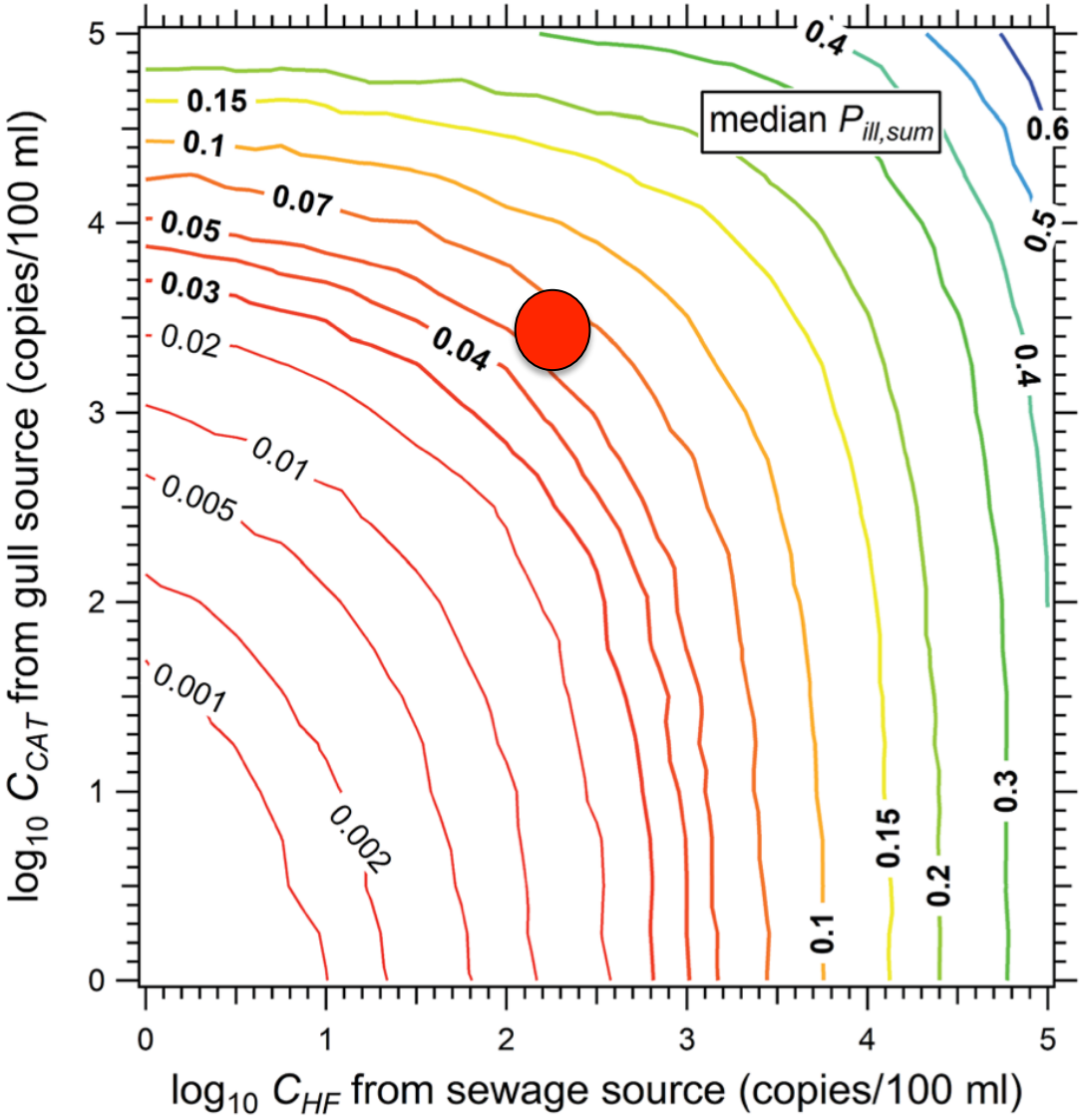
Mixture scenario



Risk as a function of CAT and HF183



Risk as a function of CAT and HF183



What if the fecal source is aged?

- If MST markers and pathogens decay at the same rate in the environment, then no change
- If MST markers decay more quickly than pathogens, then model will underestimate risk
- If MST markers decay more slowly than pathogens, then model will overestimate risk

We are currently conducting a systematic review of decay rates of human markers and QMRA pathogens to consider aging of contamination in this analysis.

Summary

MST marker (source)	Risk-based threshold (copy/100 mL)
HF183 (raw sewage)	4200
HumM2 (raw sewage)	2800
HF183 (treated effluent)	20000
CAT (gull feces)	7000
HF183 (raw sewage) & CAT (gull feces)	$\log_{10} \text{HF} = 2.95 + \frac{-2.85}{(\log_{10} C_{\text{CAT}} - 4.55)^2 + 0.26}$

Thresholds are based on the best available information and consider uncertainty and variability in the input parameters by using Monte Carlo simulations.

Your input needed

1) Do you have suggestions for future work?
2) Would you use these risk-based thresholds for MST markers for interpreting results at your beaches?