

Electronic Supplementary Information for:  
Estimating probability of illness due to swimming in  
recreational water with a mixture of human- and  
gull-associated microbial source tracking markers

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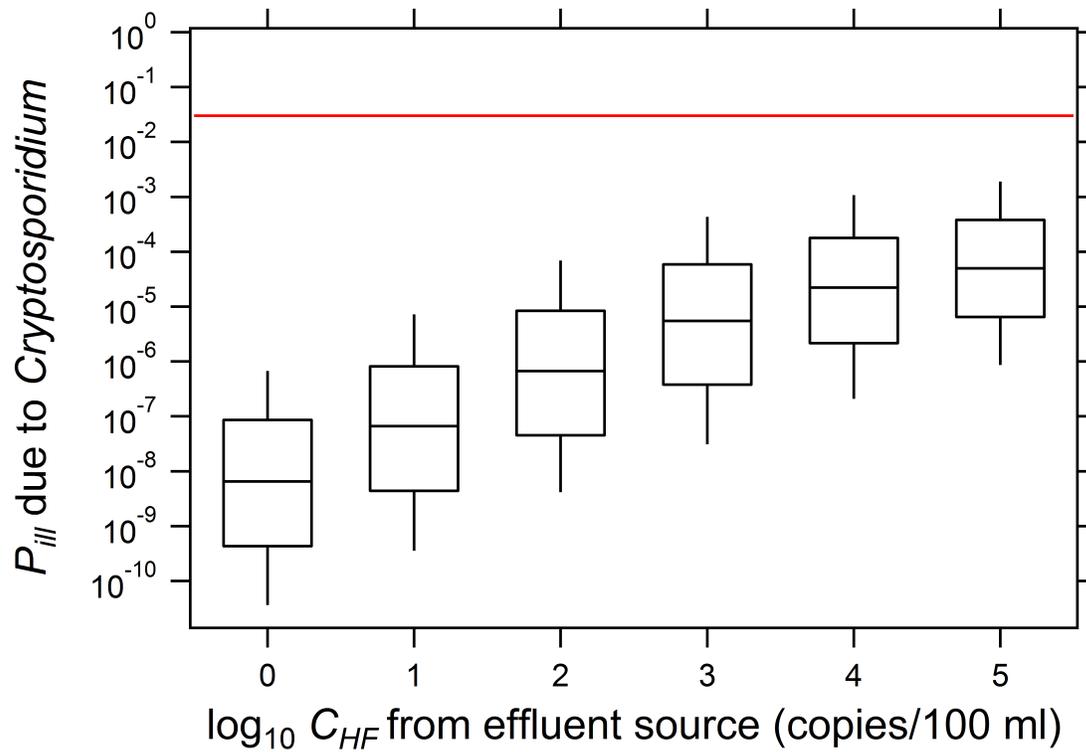


Figure S1: Box and whisker plots of  $P_{ill}^{effluent}$  due to *Cryptosporidium* versus  $\log_{10}C_{HF}$  from disinfected effluent. See Figure 2 for description. See main text for abbreviation descriptions.

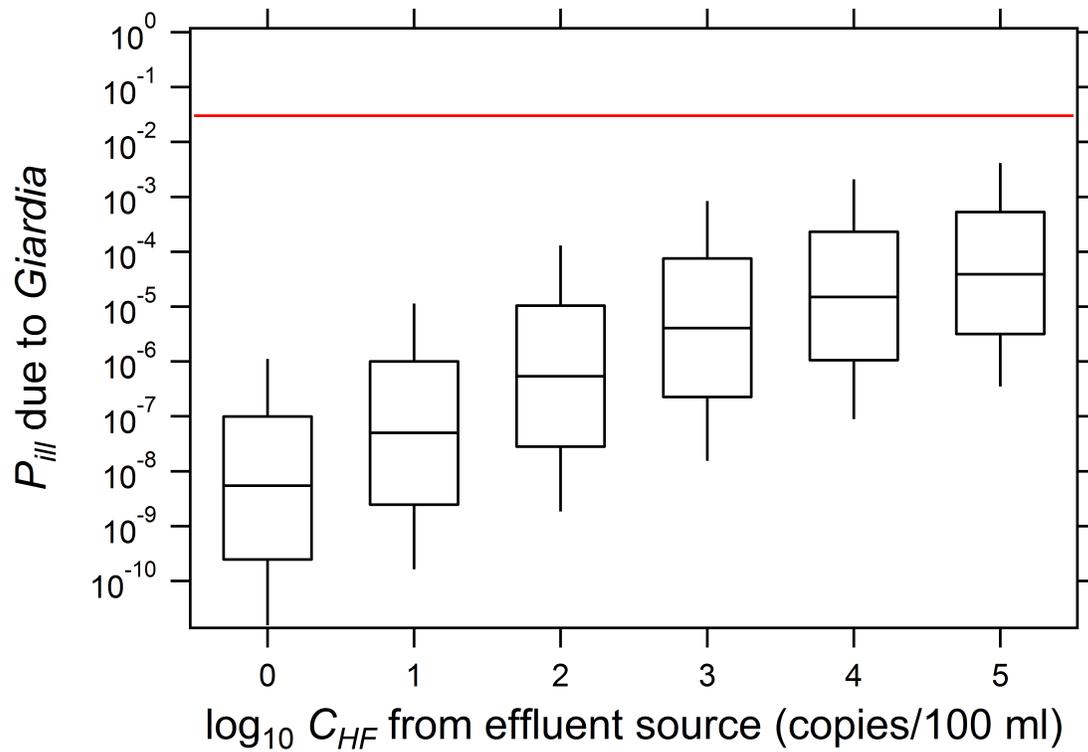


Figure S2: Box and whisker plots of  $P_{ill}^{effluent}$  due to *Giardia* versus  $\log_{10}C_{HF}$  from disinfected effluent. See Figure 2 for description. See main text for abbreviation descriptions.

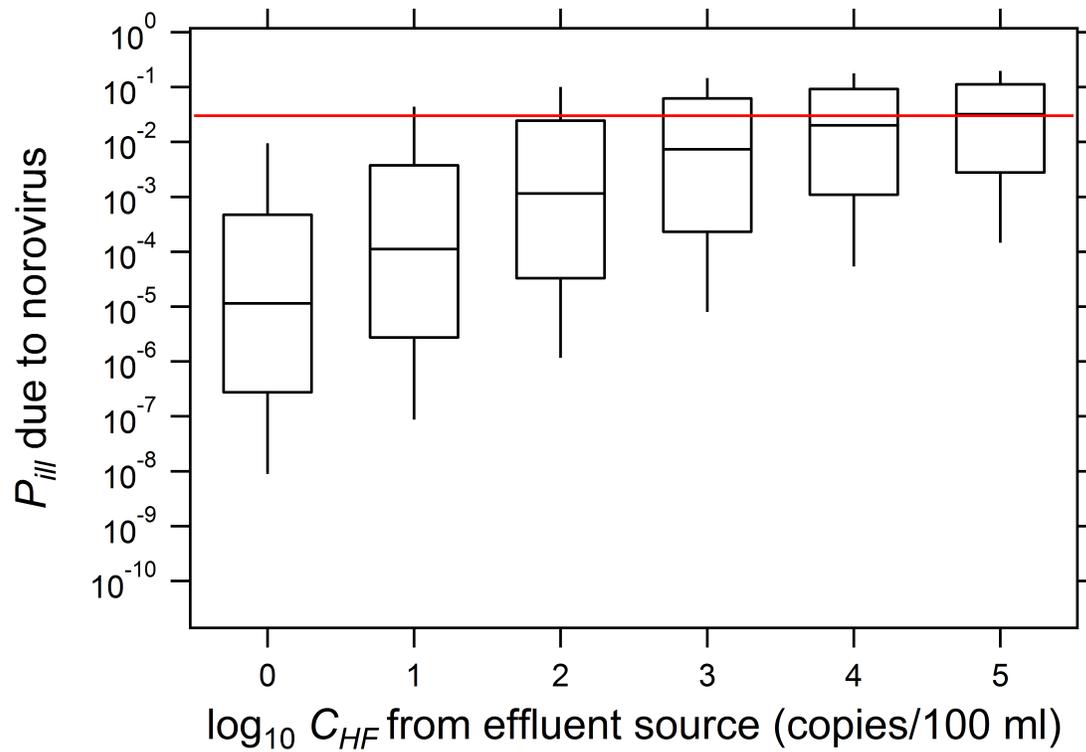


Figure S3: Box and whisker plots of  $P_{ill}^{effluent}$  due to norovirus versus  $\log_{10} C_{HF}$  from disinfected effluent. See Figure 2 for description. See main text for abbreviation descriptions.

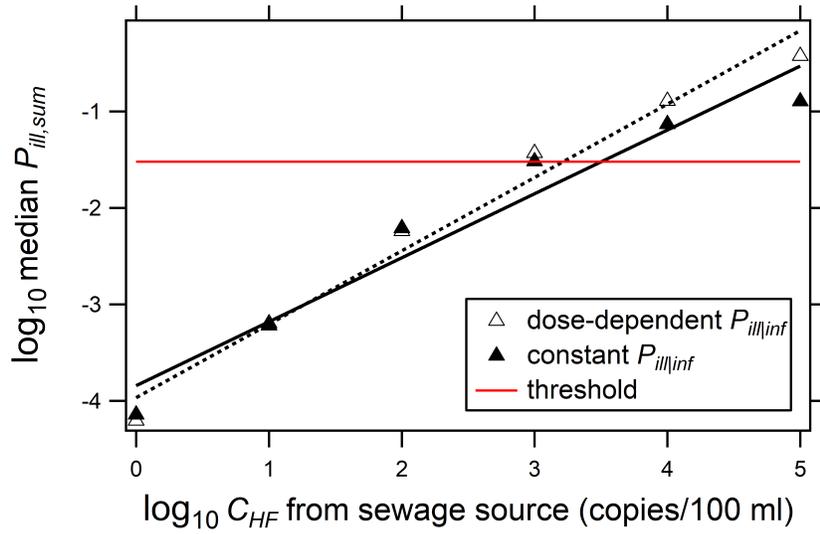


Figure S4: Median  $P_{ill,sum}^{sewage}$  at different concentrations of the HF183 human marker ( $C_{HF}^{sewage}$ ) in recreational water.  $P_{ill,sum}$  is predicted from dose-dependent versus constant  $P_{ill|inf,rp}$  for *Salmonella* and *Campylobacter*. Each  $P_{ill|inf,rp}$  is paired with a specific  $P_{inf,rp}$ ; constant  $P_{ill|inf,rp}$  with a beta-Poisson  $P_{inf,rp}$ , and dose-dependent  $P_{ill|inf,rp}$  with a beta-binomial  $P_{inf,rp}$  (Table 2). The red line indicates the threshold of 3 cases of illnesses/100 swimmers.

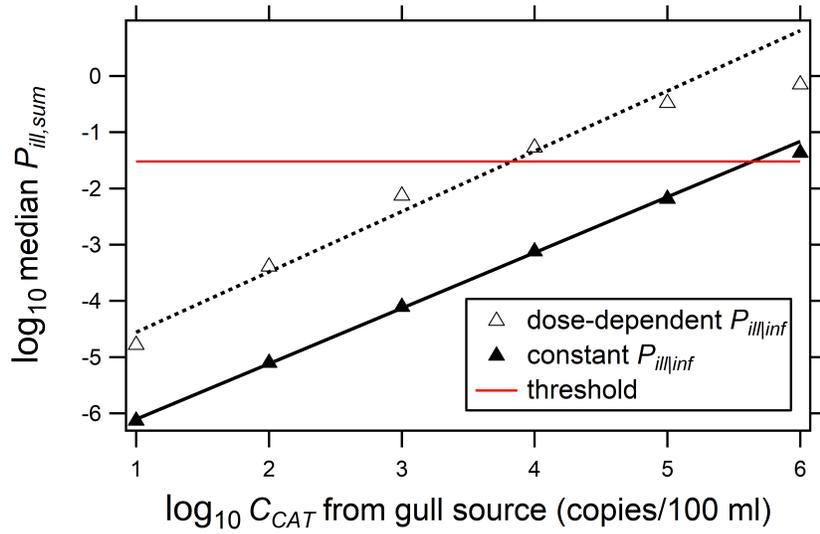


Figure S5: Median  $P_{ill,sum}^{gull}$  at different concentrations of the CAT gull marker ( $C_{CAT}$ ) in recreational water.  $P_{ill,sum}$  is predicted from dose-dependent versus constant  $P_{ill|inf,rp}$  for *Salmonella* and *Campylobacter*. Each  $P_{ill|inf,rp}$  is paired with a specific  $P_{inf,rp}$ ; constant  $P_{ill|inf,rp}$  with a beta-Poisson  $P_{inf,rp}$ , and dose-dependent  $P_{ill|inf,rp}$  with a beta-binomial  $P_{inf,rp}$  (Table 2). The red line indicates the threshold of 3 cases of illnesses/100 swimmers.

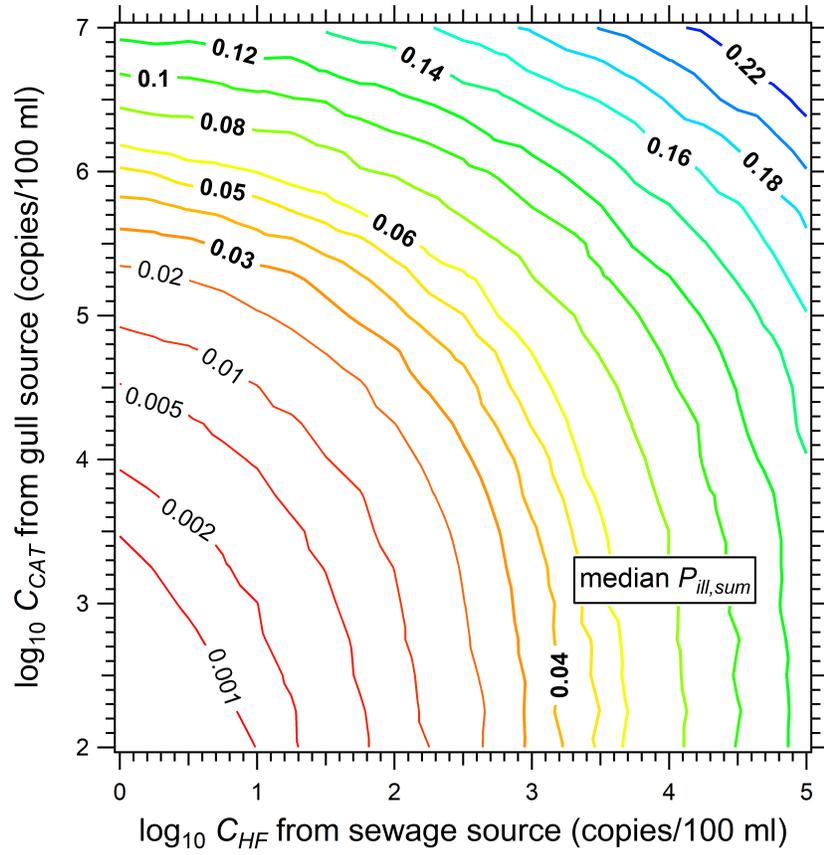


Figure S6: Median  $P_{ill,sum}$  at different mixed concentrations of the HF183 human marker ( $C_{HF}^{sewage}$ ) and the gull marker ( $C_{CAT}$ ) in recreational water. Data in this plot were generated with a QMRA using the beta-Poisson dose response curves ( $P_{inf,rp}$ ) and constant  $P_{ill|inf}$  given in Table 2.

Table S1: Primer and probe sequences used to target segments of human *Bacteroides* 16S rRNA by the HF183 qPCR assay.

Primer/ Probe	Sequence (5' to 3')	Reference
HF183	ATCATGAGTTCACATGTCCG	Bernhard and Field <sup>1</sup>
BacR287	CTTCCTCTCAGAACCCCTATCC	Green et al. <sup>2</sup>
BacP234 MGB	[6FAM]-CTAATGGAACGCATCCC-[MGB]	Green et al. <sup>2</sup>

Table S2: HF183 assay standard curve parameters. Assay performance was judged based on efficiency, with efficiency between 0.90 and 1.10 considered acceptable.

Slope	Intercept	$R^2$	Efficiency
-3.5	37.9	0.99	0.94

Table S3: Sensitivity analysis of input variables for the effluent scenario.  $F_{HF}$ : concentration of HF in effluent [copies/ml];  $L_{norovirus}$ : removal of norovirus during wastewater treatment;  $V$ : volume of seawater ingested [ml];  $R_{norovirus}$ : concentration of norovirus in sewage [genomes/L];  $R_{Giardia}$ : concentration of *Giardia* in wastewater effluent [CFU/L];  $R_{Cryptosporidium}$ : concentration of *Cryptosporidium* in wastewater effluent [CFU/L];  $P_{ill|inf,norovirus}$ : probability of illness given infection with norovirus;  $P_{ill|inf,Cryptosporidium}$ : probability of illness given infection with *Cryptosporidium*;  $P_{ill|inf,Giardia}$ : probability of illness given infection with *Giardia*.

Variable	Input Value			p75 : p25					
	p25	p50	p75	10 <sup>0</sup>	10 <sup>1</sup>	10 <sup>2</sup>	10 <sup>3</sup>	10 <sup>4</sup>	10 <sup>5</sup>
(copies HF/100 ml ambient seawater)									
$R_{norovirus}$	$4.9 \times 10^3$	$5.0 \times 10^4$	$5.1 \times 10^5$	104	102	89.5	40.2	7.21	1.84
$F_{HF}$	$1.3 \times 10^2$	$1.1 \times 10^3$	$1.2 \times 10^4$	95.4	94.2	83.9	40.4	7.6	2.0
$L_{norovirus}$	$9.4 \times 10^1$	$7.1 \times 10^2$	$5.3 \times 10^3$	55.9	55.3	4.57	25.9	5.39	1.62
$P_{ill inf,norovirus}$	0.425	0.55	0.675	1.59	1.59	1.59	1.59	1.59	1.58
$V$	13.1	14.0	15.0	1.15	1.15	1.15	1.13	1.06	1.01
$R_{Giardia}$	0.531	5.62	59.6	1.0	1.0	1.0	1.0	1.01	1.05
$R_{Cryptosporidium}$	0.266	1.41	7.50	1.0	1.0	1.0	1.0	1.0	1.03
$P_{ill inf,Cryptosporidium}$	0.325	0.45	0.575	1.0	1.0	1.0	1.0	1.0	1.0
$P_{ill inf,Giardia}$	0.325	0.45	0.575	1.0	1.0	1.0	1.0	1.0	1.0

## 16 **References**

- 17 [1] Bernhard, A. E.; Field, K. G. A PCR assay to discriminate human and ruminant feces on  
18 the basis of host differences in Bacteroides-Prevotella genes encoding 16S rRNA. *Applied and*  
19 *Environmental Microbiology* **2000**, *66*, 4571–4574.
- 20 [2] Green, H. C.; Haugland, R. A.; Varma, M.; Millen, H. T.; Borchardt, M. A.; Field, K. G.;  
21 Walters, W. A.; Knight, R.; Sivaganesan, M.; Kelty, C. A.; Shanks, O. C. Improved HF183  
22 quantitative real-time PCR assay for characterization of human fecal pollution in ambient sur-  
23 face water samples. *Applied and Environmental Microbiology* **2014**, *80*, 3086–3094.