Persistence and Decay of Fecal Microbiota in Aquatic Habitats

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SUPPLEMENTAL MATERIALS

Table S1. Effect of sunlight on decay of indicator microorganisms and pathogens in marine and freshwater. + denotes greater decay rate in the presence of sunlight; 0 denotes no effect of sunlight on decay rate.

Organism	Effect of sunlight on decay	Spike Source	Water Type	Method	Comment	Other factors contributing to decay	Reference
FIB							
Fecal coliforms	+ [b]	Raw sewage and effluent from meat-processing facility	Seawater	Membrane filtration on mE/esculin-iron agar (enterococci) or mFC agar (fecal coliforms)	Field study. Fecal coliforms inactivated more rapidly compared to enterococci	Temperature	(1)
Enterococci	+ [b]						
Fecal coliforms	+ [b]	Raw sewage	Seawater	Membrane filtration on mFC agar	Field study. Fecal coliforms inactivated more rapidly compared to coliphages	Temperature, seasonality	(2)
Enterococci	+ [b]	Waste stabilization pond	Freshwater (river)	Membrane filtration on mE/esculin-iron agar (enterococci) or mFC agar/nutrient agar supplemented with MUG (*E. coli*)	Field study. Enterococci and *E. coli* inactivated more rapidly compared to coliphages	Seasonality, salinity	(3)
E. coli	+ [b]						

E. coli	+ [a]	Raw sewage, final sewage effluent	Seawater and freshwater (creek)	Colilert (MPN)	Field study. Enterococci decayed significantly faster compared to *E. coli*	Temperature	(4)
Enterococci	+ [a]			Enterolert (MPN)			
E. coli	+ [b]	Waste stabilization pond	Seawater and freshwater (river)	Membrane filtration on mFC agar/nutrient agar supplemented with MUG (*E. coli*)	Field study. Faster inactivation in seawater compared to freshwater	Seasonality	(5)
Enterococci	+ [a]	Human, cattle and dog feces	Seawater	Membrane filtration on mEI (enterococci) and by qPCR (Entero1a)	Field study. Intact cells (as determined by the PMA treatment) and DNA persisted longer	None reported	(6)
Enterococcus qPCR	0 [a]						
E. coli	+ [a]	Human and cattle feces	Freshwater (river)	Colilert (MPN)	Field study. Exposure to sunlight affected culturable *E. coli* from cattle feces, but not human.	None reported	(7)
Enterococci	0 [b]			Enterolert (MPN)			

E. coli	+ [a]	Raw sewage	Freshwater (river)	Colilert (MPN)	Lab study. Culturable *E. coli* inactivated more rapidly than any of the qPCR markers tested	Biotic interactions, sediment	(8)
Enterococci	+ [b]	Raw sewage	Seawater and freshwater (creek)	Enterolert (MPN) and by qPCR (Entero1a)	Field study. Decay rate of DNA significantly lower compared to culturable enterococci.	Water type	(9)
Enterococcus qPCR	0 [b]						
E. coli	+ [a]	Strains isolated from cattle manure	Freshwater (pond)	Colilert (MPN)	Field study. Effect of sunlight appeared to be seasonal (not observed during the winter months)	Temperature, biotic interactions	(10)
Enterococci	+ [a]			Enterolert (MPN)			
E. coli	+ [b]	Human, cattle and dog feces	Groundwater	Membrane filtration on mEI (enterococci) and by qPCR (Entero1a), membrane filtration on mTEC (*E. coli*)	Field study. Intact cells (as determined by the PMA treatment) and DNA persisted similarly in light and dark treatments	None reported	(11)
Enterococci	0 [a]						
Enterococcus qPCR	0 [a]						
E. coli	0 [a]	Raw sewage and cattle feces	Freshwater (lake)	Colilert (MPN)	Field study. Water temperature had	Temperature	(12)
Enterococci	0 [a]			Membrane filtration on Slanetz& Bartley			

				agar, followed by Bile Esculin Azide Agar	significant impact on decay		
Enterococci	+ [a]	Laboratory grown strain	Seawater	Enterolert (MPN), membrane filtration on mEI, spread plating on TSA and by qPCR (Entero1a) with and without PMA	Field study. Under anoxic conditions, sunlight did not affect decay of culturable enterococci when enumerated by Enterolert and TSA, but it was a significant factor when enterococci when enumerated using mEI	Oxidative stress	(13)
Enterococcus qPCR	0 [a]						
Enterococci	+ [a]	Cattle manure, primary treated sewage	Seawater and freshwater (river)	Membrane filtration on mEI (enterococci) and mTEC (*E. coli*)	Field study. Sunlight significantly affected only decay of sewage-borne enterococci.	Biotic interactions, fecal source	(14)
E. coli	0 [a]						
Enterococci	+ [a]	Primary treated sewage	Freshwater (river)	Membrane filtration on mEI (enterococci) and by qPCR (Entero1a)	Field study. Sunlight exposure was more important for culturable *E. coli* compared to enterococci. There was no statistically significant correlation in decay of culturable enterococci compared	Biotic interactions	(15)
Enterococcus qPCR	+ [a]						
E. coli	+ [a]			Membrane filtration on mTEC			

					to the corresponding qPCR signal.		
Enterococci	+ [a]	Raw sewage, human feces	Seawater	Membrane filtration on mEI (enterococci) and by qPCR (Entero1a)	Field study. Culturable enterococci and *E. coli* decayed faster than their molecular counterparts and were affected by sunlight more.	Biotic interactions, fecal source	(16)
Enterococcus qPCR	+ [a]						
E. coli	+ [a]			Membrane filtration on mTEC and by qPCR (EC23S857)			
E. coli qPCR	+ [a]						
Enterococci	+ [b]	Raw sewage	Seawater, brackish water, freshwater (lagoon)	Enterolert (MPN)	Field study. Generally faster decay in clear (seawater) and shallow waters.	None reported	(17)
E. coli	+ [b]			Colilert (MPN)			
Enterococci	+ [a]	Raw sewage	Seawater	Enterolert (MPN) and by qPCR (Entero1a)	Field study. Enterococci (culture and qPCR) decayed faster in summer than in the winter under the same sunlight intensity, while the opposite was the case for *E. coli*.	Seasonality	(18)
Enterococcus qPCR	+ [a]						
E. coli	+ [a]			Colilert (MPN)			

E. coli qPCR	0[a]	Cattle feces	Freshwater (river) and seawater	qPCR	Field study. *Enterococcus* qPCR signal decayed significantly faster than *E. coli*. Culturable enterococci and *E. coli* decayed faster than their molecular counterparts.	Water type	(19)
Enterococcus qPCR	0[a]						
Bacterial Pathogens							
Salmonella enterica	+ [b]	Laboratory grown strains	Seawater and freshwater (river)	Preston broth/ Exeter agar (MPN, *Campylobacter*), membrane filtration on XLD agar (*Salmonella)*	Field study. *Campylobacter* and *Salmonella* inactivated more rapidly compared to *E. coli*	Seasonality	(5)
Campylobacter jejuni	+ [b]						
E. coli O157:H7	+ [b]	Laboratory grown strain	Freshwater (pond)	Lauryl tryptose broth (LTB) enrichment combined with qPCR for confirmation (MPN)	Field study. Decayed significantly slower compared to FIB	None reported	(10)
Salmonella enterica	0 [a]	Laboratory grown strains	Groundwater	qPCR	Field study. Intact cells (as determined by the PMA treatment) but not DNA (no PMA) of *C. jejuni* decayed faster when exposed to	None reported	(11)
Campylobacter jejuni	0 [a]						

					sunlight compared to dark treatments.		
Campylobacter jejuni	+[b]	Laboratory grown strains	Freshwater (river)	Preston broth enrichment, followed by sub-culturing on Karmali agar (MPN)	Lab and field study. No correlation between decay and pH, oxygen concentrations or conductivity	Temperature	(20)
Coliphage							
Somatic coliphage	+ [b]	Raw sewage	Seawater	Double agar layer and/or membrane filtration-swirling elution method	Field study. F-RNA coliphage more susceptible to longer solar wavelengths than somatic coliphage	Temperature, seasonality	(2)
F-RNA coliphage	+ [b]						
Somatic coliphage	+ [b]	Waste stabilization pond	Freshwater (river)	Double agar layer and/or membrane filtration-swirling elution method	Field study. Somatic coliphage persisted longer than F-RNA coliphage	Seasonality, salinity	(3)
F-RNA coliphage	+ [b]						
F-specific coliphage	+[a]	Raw sewage, final sewage effluent	Seawater and freshwater (creek)	Double agar layer	Field study. Coliphage decayed significantly slower than FIB	Temperature	(4)
Somatic coliphage	+ [b]	Laboratory grown strains	Seawater	Double agar layer	Lab study. Somatic coliphage more sensitive to light	None reported	(21)
F-specific coliphage	+ [b]						

					compared to F-RNA coliphage		
Somatic coliphage	0 [a]	Raw sewage and cattle feces	Freshwater (lake)	Double agar layer	Field study. Persistence significantly lower in August compared to March and November	Temperature	(12)
F-specific coliphage	+ [b]	Laboratory grown strain	Seawater, brackish, freshwater (lagoon, wetland)	Double agar layer	Lab study. Exogenous sunlight damage caused by external reactive species was more important than the direct (endogenous) damage	Water composition	(22)
F-specific coliphage	+ [b]	Laboratory grown strain	Freshwater (wetland)	Double agar layer	Lab and field study. No significant difference in decay at two different depths (5 cm and 20 cm)	None reported	(23)
Somatic coliphage	+ [a]	Raw sewage	Seawater	Double agar layer	Field study. Somatic coliphage more sensitive to light compared to F-specific coliphage	Biotic interactions	(16)
F-specific coliphage	0 [a]						
Viral Pathogens							
Adenovirus 2	+ [a]		Seawater				(21)

Poliovirus 3	+ [a]	Laboratory grown strains		Mammalian cell culture	Lab study. MS2 coliphage and Adenovirus2 more resistant to sunlight compared to other coliphages and Poliovirus	None reported	
Adenovirus 2	0 [a]	Laboratory grown strain	Ground water	qPCR	Field study. No significant difference in decay compared to *C. jejuni* and *S. enterica* with respect to dark and sunlight exposed treatments	None reported	(11)
Poliovirus 3	+ [b]	Laboratory grown strain	Seawater, brackish, freshwater (lagoon, wetland)	Mammalian cell culture	Lab study. Generally, Poliovirus decayed faster than Adenovirus or F-specific (MS2) coliphage	Water composition	(22)
Adenovirus 2	+ [b]						
Poliovirus 3	+ [b]	Laboratory grown strain	Freshwater (wetland)	Mammalian cell culture	Lab and field study. Inactivated slower compared to the F-specific (MS2) coliphage	None reported	(23)
Protozoan Pathogens							

C. parvum	+[a]	Cattle feces	Freshwater (lake, reservoir)	Mammalian cell culture combined with qPCR	Field study. Increased dissolved organic carbon reduced inactivation	Water type	(24)
MST Markers							
Human-associated (HF183, HF134)	0 [a]	Human and cattle feces	Freshwater (river)	qPCR	Field study. Persistence of cells (as determined by RNA quantification) was significantly affected by sunlight only for the human-associated MST markers.	None reported	(7)
Ruminant-associated (CF193)	0 [a]						
Ruminant-associated (CF128)	+ [a]						
General marker of fecal pollution (BacUni-UCD)	0 [a]	Human, cattle and dog feces	Seawater	qPCR	Field study. Detection of intact cells (as determined by the PMA treatment) and DNA differed significantly in both light exposed and dark treatments	None reported	(6)
Human-associated (BacHum-UCD)	0 [a]						
Dog-associated (BacCan-UCD)	0 [a]						
Cow-associated (BacCow-UCD)	+ [a]						
General marker of fecal pollution (AllBac)	+ [a]	Raw sewage	Freshwater (river)	qPCR	Lab study. Decay of human-associated	Biotic interactions, sediment	(8)

Human-associated (HF183 and BacHum)	+ [a]				MST markers was similar		
General marker of fecal pollution (GenBac3)	0 [a]	Raw sewage	Seawater and freshwater (creek)	qPCR	Field study. Persistence longer in seawater compared to freshwater	Water type	(9)
Human-associated (BsteriF1, BuniF2, HF183, HF124, HumM2)	0 [a]						
General marker of fecal pollution (BacUni-UCD)	0 [a]	Human, cattle and dog feces	Groundwater	qPCR	Field study. Intact (PMA) treated cells persisted significantly longer than the (non-PMA treated) DNA. Cow associated MST marker decayed more rapidly than others.	None reported	(11)
Human-associated (BacHum-UCD)	0 [a]						
Dog-associated (BacCan-UCD)	0 [a]						
Cow-associated (BacCow-UCD)	0 [a]						
Human-associated (BacH)	0 [a]	Raw sewage and cattle feces	Freshwater (lake)	qPCR	Field study. Persistence of MST markers not significantly different from FIB and coliphage	Temperature	(12)
Ruminant associated (BacR)	0 [a]						

General marker of fecal pollution (GenBac3)	+[a]	Primary treated sewage	Freshwater (river)	qPCR	Field study. More pronounced effect of sunlight in the early stages of decay (< 72 hours) compared to subsequent time points	Biotic interactions	(15)
Human-associated (HF183 and HumM2)	+[a]						
General marker of fecal pollution (GenBac3)	0[a]	Cattle feces	Freshwater (river) and seawater	qPCR	Field study. Sunlight affected the decay of CowM2 and Rum2Bac, but not CowM3	Water type	(19)
Cattle/ruminant associated (CowM2, CowM3, Rum2Bac)	+/0[a]						
Human-associated (HF183 and HumM2)	+[a]	Raw sewage, human feces	Seawater	qPCR	Field study. The magnitude of the effect varied across different time points and pollution sources	Biotic interactions, fecal source	(16)
General marker of fecal pollution (GenBac3)	+[a]						
General marker of fecal pollution (GenBac3)	0[a]	Raw sewage	Seawater	qPCR	Field study. When exposed to sunlight, culturable FIB decayed significantly faster than the MST markers	None reported	(18)
Human-associated (HF183, HumM2 and BacHum)	0[a]						

[a]Statistical significance reported

[b]Statistical significance not reported

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