Topanga State Beach
Microbial Source Tracking Study

Conducted by University of California Los Angeles (UCLA)[1] in collaboration with the Resource Conservation District of the Santa Monica Mountains (RCDSMM) and the Southern California Coastal Water Research Project Authority (SCCWRP) as part of the Source Identification Protocol Project (SIPP)

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<u>Disclosure statement</u>

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Contents

Background

Topanga State Beach was selected for a microbial source tracking investigation as part of SIPP. This beach was included as a field site because it continues to receive poor water quality ratings. Topanga State Beach is frequently listed as one of the most impacted beaches in the state of California (Heal the Bay 2013) based on fecal indicator bacteria (FIB) levels. Ranked 9th most polluted California beach in 2006, and 4th in 2011, Topanga State Beach has FIB exceedances well into the summer season.[1] In addition, Topanga Creek was listed by the Regional Water Quality Control Board 303(d) for bacteria at Topanga State Beach. The last comprehensive sampling and monitoring in the Topanga Creek Watershed took place in 2003-2004. This sampling effort revealed that 50% of samples collected during storm events were in exceedance for enterococci and two samples collected from Topanga Creek were positive for enterovirus.[2] A review of historical data taken by the Los Angeles County Department of Public Health revealed that when these data were compared to creek flow data collected by the County at the same time as the bacterial data, bacterial exceedances at the beach correlated strongly with breaches in the Topanga Lagoon. Over the years analyzed, the Topanga Lagoon discharged episodically into the ocean as late as July. This correlation between Lagoon discharges and high FIB values in ocean water samples strongly suggested that Topanga Lagoon was a primary source of bacterial pollution to the ocean.

Topanga State Beach is located in Los Angeles County, California and receives an estimated 750,000 annual visitors. Topanga Creek watershed (approximately 47 km^2) is 70% undeveloped and includes a creek and lagoon system[3] (Figure 1). Topanga Creek drains the upper watershed and culminates in Topanga Lagoon, a dynamic lagoon system that breaches and berms throughout the year, contributing variable flow to \the beach. The lagoon contains brackish water and houses a large population of gulls and other waterfowl. Dwellings and businesses in the watershed are not connected to a centralized sanitary sewer system. Instead, sanitary waste is disposed onsite utilizing septic systems. An exception are businesses located adjacent to the lagoon which have holding tanks that are pumped out on a regular basis.

Findings of the Topanga State Beach microbial source tacking study may apply to other beaches with similar creek and lagoon systems. These results may help inform improved sampling strategies and design in future MST studies, thereby allowing for better identification of problem areas requiring immediate remediation efforts.

Potential Sources

Sources can be divided into two categories, lower watershed sources and upper watershed sources that travel to the beach via the creek.

Potential Lower Watershed Sources

1. *Septic tanks along Pacific Coast Highway in Topanga State Park.* The septic tanks at Cholada's restaurant, Ranch Motel Ranger residence, Reel Inn, Malibu Feed Bin, and the new winery are pumped weekly or more as needed to comply with contracts administered by the California Department of Parks and Recreation. These old septic tanks supposed to be disconnected from leach fields or seepage pits, but the potential for leakage is present.

[1] Heal the Bay, via http://brc.healthebay.org
[2] Dagit, R., Krug, J., Adamek, K., et al. 2013. Topanga Source ID Study Annual Report 2012 - 2013 Report. Resource Conservation District of the Santa Monica Mountains, Topanga, CA.
[3] GeoPentech, 2006. Hydrogeologic Study, Lower Topanga Creek Watershed, Los Angeles County. California. Resource Conservation District of the Santa Monica Mountains.

Beaches and Harbors restrooms and lifeguard station. While a stand-alone treatment facility exists at this site, it needs to be evaluated and listed as a potential source in the event of a malfunction and/or excluded as a potential source.

2. *Wildlife, including gulls and other seabirds, deer, coyotes.* Although it is only 1.8 acres, the remnant lagoon at the mouth of Topanga Creek is consistently used by roosting and foraging waterfowl. Bacterial contributions from bird feces have been identified as the source of exceedances in other coastal lagoons, such as Cowell Beach in Santa Cruz[4] and a beach in Racine, WA[5].

Potential Upper Watershed Sources

1. *Homes on septic systems throughout watershed.* Many homes in Topanga were built in the 1920's and 1930's and the septic systems are old. Approximately 200 of the 3,000 homes in the watershed are built directly adjacent to the creek.
2. *Transient encampments.* Homeless persons frequently inhabit several locations throughout the watershed. While encampments are dispersed whenever identified, it is possible that new encampments exist.
3. *Horses.* There are several establishments housing large numbers of horses in the watershed and many residents have one or two horses on fairly small parcels. Horses are ridden in open land throughout the watershed, resulting in a potentially diffuse bacterial source. Horse feces at barns are sometimes composted and these piles could serve as a bacterial source.
4. *Dogs.* Fecal matter from the many household dogs may be a potential diffuse source of bacteria in the watershed.
5. *Wildlife including coyotes, deer and birds.* 70% of the watershed is undeveloped; thus, the watershed is home to coyotes, deer, native pond turtles, mountain lions, and other species.

Stakeholder involvement

A Technical Advisory Committee (TAC) was convened to provide oversight and assist in fine-tuning the sampling design and analysis for the project. The TAC is comprised of stakeholder representatives including Los Angeles County Departments of Beaches and Harbors, Public Health, Public Works and the Third District Supervisorial Office. Non-county affiliated members include the California Department of Parks and Recreation, Caltrans, Los Angeles Regional Water Quality Control Board, BioSolutions, Topanga Underground, as well as scientists from the Southern California Coastal Water Research Project (SCCWRP), and the Resource Conservation District of the Santa Monica Mountains.

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[4] Russell, T. L., L. M. Sassoubre, D. Want, et al. 2013. A coupled modeling and molecular biology approach to microbial source tracking at Cowell Beach, Santa Cruz, CA, United States. *Environmental Sci. Technol.* 47, 10231–10239.

[5] Converse, R. R., J. L. Kinzelman, E. A. Sams, et al. 2012. Dramatic improvements in beach water quality following gull removal. *Environ. Sci. Technol.* 46, 10206– 10213.

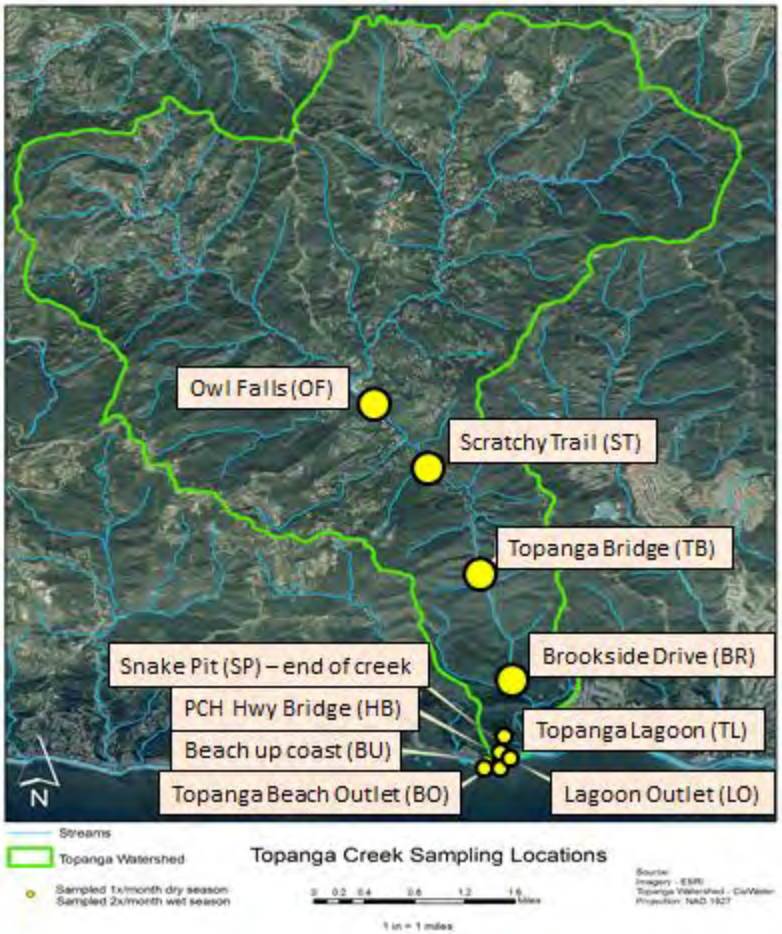

Figure 1. Map of the Topanga Creek Watershed and sampling locations. Five samples were collected from the lower half of the watershed along the main stem of Topanga Creek, three samples were collected within the lagoon and two marine samples from Topanga State Beach.

Hypotheses

Based on existing data and the possible upper and lower watershed sources described above, we developed a series of hypotheses. Sub-hypotheses were intended to be evaluated sequentially, depending on whether a previous, relevant sub-hypothesis was accepted or rejected. The phrase "tidal prism" in this context refers to the portion of foreshore sand that is washed with ocean water each tidal cycle. (Note that this is not the conventional definition of tidal prism, but we retain that wording here for consistency with the Study Plan.)

H1: The creek outlet serves as a source of FIB to the beach.

- H1.1: Topanga Creek is a major source of FIB to Topanga Beach
- H1.2: The lagoon plays a role in the enhancement of FIB levels, either by regrowth or other inputs
- H1.3: A spatial pattern exists in the lagoon that may indicate a point source.
- H1.4: Gulls are a major source of FIB to the lagoon.
- H1.5: The Upper Watershed inoculates the lagoon.
- H1.6: Sources in the Upper Watershed result in the transport of FIB to the Lower Watershed.

H2: There are sources in the tidal prism that result in elevated FIB in surf zone water.

- H2.1: Major sources of FIB are present in the tidal prism.

H2.1.1: FIB concentrations correlate to tidal cycle over 24-hour periods
H2.1.2: FIB concentrations correlate to tidal range over 1 month
H2.2: Sand, wrack etc are harboring FIB along the coast.
H2.3: Coastal FIB communicates with ocean only at furthest tidal reach
H2.4: Diffuse coastal source (bird, dog etc)

Preliminary Sampling

To identify the most relevant hypotheses for further study, we conducted preliminary "snapshot" monitoring to gather more information on various potential sources. Six snapshot sampling events occurred over a 10-month period beginning 5 October 2011. The first of these snapshots occurred during a "first flush" event, where the watershed had just received the first rain of the season. Each snapshot focused on a different study area and/or season. The sampling area and number of sites for each snapshot was as follows: Full watershed during first flush, 9; full watershed during autumn dry weather, 10; beach transect, 7; beach/lagoon, 10; full watershed during spring dry weather, 14; lagoon, 8. These samples were analyzed for FIB as well as human, dog, and gull-specific qPCR markers. Snapshot sampling indicated there were "hotspots" of FIB and human markers in the lagoon and near the town of Topanga in the upper watershed. In addition, the lagoon contained very high levels of gull markers and somewhat lower levels of dog markers. A single snapshot indicated that the high microbial loading from the town was attenuated downstream. Concentrations of indicators and markers at the beach seemed related to those in the lagoon, but not enough samples were taken to have statistical confidence in this relationship.

Revised Hypotheses

After analyzing the data from the snapshot sampling, we revised our hypotheses to further investigate the spatial and temporal patterns we observed.

H1: Topanga town is a substantial source of microbial contamination to Topanga Creek.
H2: Upper watershed sources of FIB are not conveyed to the beach via the creek.
H2.1: Concentrations of FIB and/or host-associated markers decrease as the creek flows downstream
H3: Lower watershed and/or lagoon sources of FIB (human and non-human inputs such as gull, dog, etc.) are correlated with exceedances at Topanga Beach.
H3.1: Levels of FIB at the beach are related to gull and dog marker levels.
H3.2: FIB and human markers are leaking from faulty septic systems in the lower watershed.

Project Approach

Following the snapshot surveys, we identified sources of FIB to Topanga State Beach through a combination of long-term monitoring during wet and dry seasons and measuring a suite of qPCR markers at all sites. Spatial sampling was used to confirm whether contamination is conveyed downstream via the creek to the ocean. Salinity, temperature, pH, and conductivity measurements were collected at each site. Flow and nutrient concentrations (nitrogen and phosphorus) were evaluated when/where possible.

To evaluate H1, inputs from the town of Topanga, California were investigated over a two-week intensive sampling period from 23 May – 7 June 2012. Samples were collected on three visits over the two-week period from two tributaries draining the upper reaches of Topanga, and below the confluence in the main stem of Topanga Creek. A bracketed sampling approach was used to hone in on locations with high levels of FIB. Samples were analyzed for host-associated markers and FIB using a culture based method (IDEXX) and an adaptive field portable technique, Cov-IMS/ATP for enumeration of enterococci (ENT).

To evaluate H2 and H3, select sampling locations were chosen (based on initial snapshot sampling) for long-term monitoring over a nine-month period between 17 November 2012 and 1 July 2013 (Table 1). Sites were sampled bi-monthly during the wet weather season (November – March) and monthly during the dry weather season (April – July). We captured four storm events over the course of the study, and collected a total of 35 samples during active rainfall. In order to document input from the upper watershed (H2), samples were collected at five sites within the creek. The most upstream site Owl Falls (OF) is located nearest to the town of Topanga and lies just below the confluence of the two major tributaries draining the upper portions of the watershed. Downstream lies Scratchy Trail (ST), a remote sampling location approximately 15-minutes hike from the main road. To test H3, a lagoon transect was also sampled at three sites (Highway Bridge, Topanga Lagoon and Lagoon Outlet). Two marine samples were collected from Topanga State Beach at Beach Outlet (BO) and Beach Upper (BU). BO was sampled at the outlet of the lagoon, whether the lagoon was bermed or breached. Beach Upper (BU) was collected approximately a quarter mile north of the lagoon in the ocean, to represent marine water quality conditions upstream of the lagoon discharge point.

Table 1. Description of sampling locations (Coordinate System: UTM , Zone 11N) and sampling frequency.

Site Name	Easting (m)	Northing (m)	Elevation (ft)	Number Samples Wet Season	Number Samples Dry Season
Beach Upcoast (BU)	353726	3767515	0	2/mo + first flush	1/mo
Beach Outlet (BO)	353896	3767506	0	2/mo + first flush	1/mo
Lagoon Outlet (LO)	353872	3767529	0	2/mo + first flush	1/mo
Lifeguard Station Beach (LG)	353968	3767553	0	2/mo + first flush	1/mo
Topanga Lagoon (TL)	353887	3767573	0	2/mo + first flush	1/mo
PCH Bridge - 0m (HB)	353868	3767649	0	2/mo + first flush	1/mo
Lifeguard Station Septic (LS)	353994	3767655	0	1/mo	1/mo
Snake Pit – 300m (SP)	354015	3767841	0	2/mo + first flush	1/mo
Brookside Drive – 1700m (BR)	354075	3768713	0	2/mo + first flush	1/mo
Topanga Bridge – 3600m (TB)	353522	3770391	200	2/mo + first flush	1/mo
Scratchy Trail – 4800m (ST)	353518	3771500	500	1/mo + first flush	1/mo
Owl Falls – 6500m (OF)	352673	3772373	700	1/mo + first flush	1/mo
Falls Drive (FD)	352535	3772259	750	occasional	
Behind Abuelita's (BA)	351570	3772891	700	occasional	

Project Outcomes

Our monthly sampling survey revealed chronic “hot spots” of microbial pollution affecting Topanga State Beach. The frequency of human markers in the upper watershed suggested presence of a unique source of human fecal contamination related to septic systems or potential homeless encampments. While FIB loading to the upper watershed was substantial, upper watershed sources appear unrelated to FIB levels at the lagoon or ocean sites. A consistent decrease in indicator bacteria and source markers occurred between the upmost creek site and downstream lagoon sampling locations. Increased bacteria levels and presence of human, gull and dog-associated markers at lagoon sites suggest an independent source near the lagoon and eliminated the creek as the source of FIB exceedances at Topanga State Beach. Dog, gull and human-associated markers were found frequently in the lagoon and ocean.

Regarding H1, the sampling effort of 2012 identified a hot spot of high ENT levels and related human-associated marker in the town region of the Topanga watershed. To better understand the nature and extent of this hot spot, samples were taken three times over a two-week period in an attempt to bracket in the source. The intensive sampling with the FIB enumeration methods indicated that the FIB source was coming from both the main stem of the creek and the Old Topanga Creek tributary. These sample events were further analyzed for host-associated markers. The human and dog marker were both detected in the Old Topanga Creek tributary. The horse-associated marker was not detected at any site.

Regarding H2, FIB levels are frequently elevated in the upper watershed, particularly at Owl Falls (furthest upstream creek sample). However, except for rain events or observed transient activity, bacterial levels decreased as the flow moved downstream to the lowest creek sampling site at the Snake Pit (located 300 meters upstream from the Topanga Lagoon). Samples collected within the lagoon and the ocean had clearly different patterns than those observed upstream within Topanga Creek. Nutrient levels in Topanga creek and lagoon were overall low, and despite the very low flow conditions in 2012-2013, the pattern of decreasing levels of nutrients as the creek flows downstream were consistent with those observed in previous studies (Dagit et al. 2004). Exceptions to this pattern were observed during rain events and associated with transient activities.

Regarding H3, Beach Outlet exhibited similar FIB trends as Topanga Lagoon, but with lower concentrations. FIB levels were fairly consistent at Beach Outlet throughout the year (Figure 2). The lagoon also had consistent high levels of FIB and frequent low levels of human-associated markers. Sporadic human hits were more frequent at Beach Outlet during the winter months, with concentrations ranging up to 10^3 copies/100 mL. Presence of markers corresponded with recorded visual observations of human feces and transient activity. Homeless encampments were found and dismantled throughout the watershed; this is a continuing issue for the County. Transient activity adjacent to the lagoon (HB and TL sites) was recorded for several months (January to March 2013) and found to directly impact water quality near these sites. Stakeholder involvement within this study also enabled testing of infrastructure, which revealed two faulty septic systems adjacent to Topanga Lagoon.

Both gull and dog-associated markers were present at high frequencies and were identified as important sources to Topanga Lagoon and State Beach. Gulls and other waterfowl have been found to impair water quality at other beaches[6] and may be responsible for FIB exceedances in surf zone and lagoon samples. The dog marker was detected more frequently and at higher levels during the winter months at both the beach and the lagoon (Figure 2). This may be related to the decreased lifeguard presence at Topanga State Beach during the winter season. Current regulations prevent dogs on this beach, but a lack of enforcement may be problematic. Higher dog marker levels at BU versus BO could be due to the fact that the lifeguard patrol ends just east (downcoast) of BU, and many of the residences along the beach upcoast have dogs. Although it is difficult to control for fecal contributions from wildlife, watershed managers can mitigate fecal waste from pets. Contamination associated with domestic dogs could be reduced by increased enforcement by lifeguards, better signage, and community education and awareness.

Lessons Learned

1) FIB and host-associated marker levels appear to consistently decrease downstream from the town of Topanga. Upstream sources of FIB and markers appear to be separate from and independent of those detected downstream at the lagoon and beach.
2) Transient populations are contributors to fecal pollution at the lagoon and possibly upstream in the creek.
3) Faulty septic systems may have been a source of FIB and human-associated markers.
4) Dogs are a source of FIB to both the lagoon and ocean.
5) Gulls are a source of FIB to the lagoon and ocean.

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[6] Lu J., Santo Domingo J.W., Lamendella R., et al. 2008. Phylogenetic diversity and molecular detection of bacteria in gull feces. Appl Environ Microbiol. 74, 3969-3976.

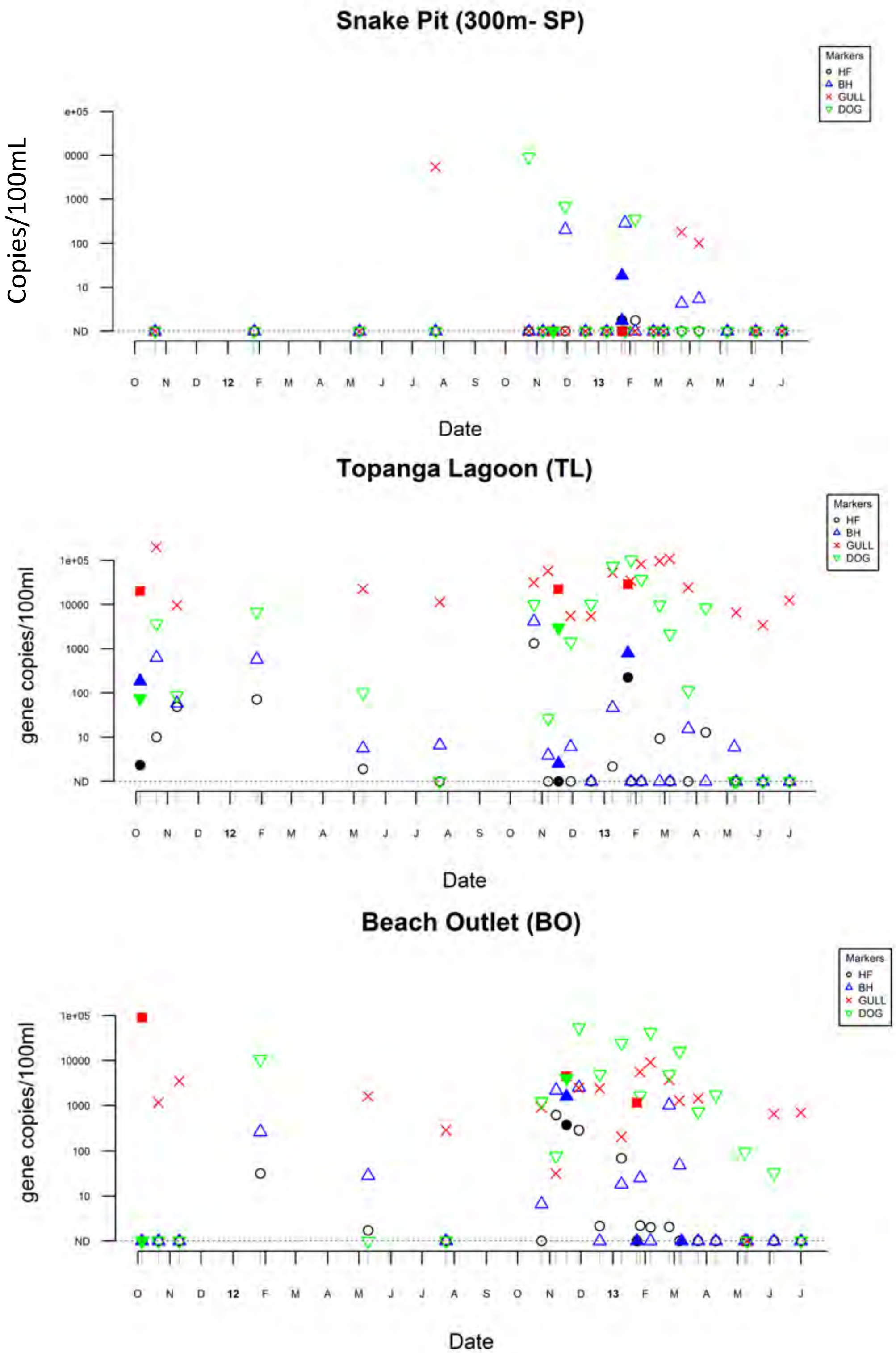

Figure 2. Time-series plots for Topanga creek (SP), lagoon (TL) and beach (BO) depict trends in host-associated markers over different seasons. Grey dottet line indicates the sample limit of detection. Concentrations in gene copies/100ml are plotted for the human-associated markers (HF183, black circle and BacHum, blue triangle), gull-associated marker (Gull, red x) and the dog-associated marker (Dog, green triangle). Note that rain events are included in this figure and indicated with solid symbols.

Next Steps

Testing of the septic systems along PCH indicated that the system at the Ranger residence at the Topanga Ranch Motel was possibly leaking: repairs were completed in summer 2013 (Figure 3). The system at the Malibu Feed Bin, a local animal feed store, was also a potential source of leachate and requires repair and further testing to evaluate the input potential into Topanga Creek. The other systems within Topanga State Park do not appear to be leaking, nor does the County Lifeguard septic system.

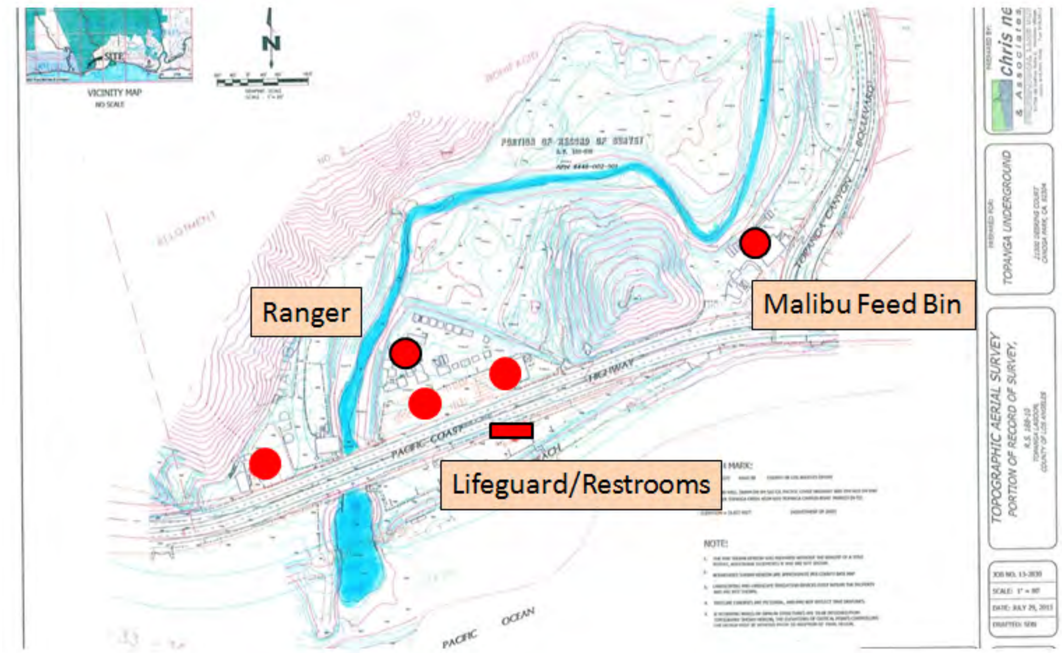

Figure 3. Aerial map of the septic systems located in Topanga State Park adjacent to the lagoon and creek system. These five systems were tested for leaks and for their potential connectivity in summer 2013.

Further, an active outreach effort providing information on the effects of dog waste on water quality is underway. UCLA will conduct a serving learning class where an emphasis will be placed on the effect of dog waste on local water quality. In addition, a community meeting is scheduled to inform local residents on the use of rainwater harvesting and cisterns to reduce run off from residential and commercial properties.

Appendix A: Technical Report

Title

Long term molecular marker monitoring at Topanga State Beach suggests that fecal indicating bacteria are from multiple animal hosts, each following different seasonal patterns.

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Abstract

High levels of fecal indicator bacteria (FIB) have been observed at Topanga State Beach resulting in exceedances of water quality standards and postings of beach advisory warnings. The origin of the bacteria impairing water quality at the beach is unknown. Analysis of historical FIB concentrations suggests Topanga Creek (which terminated in a small degraded lagoon) discharge to be the main source of bacteria to the surf zone. The Topanga watershed is primarly developed in the upper and lower watershed with the middle section consisting entirely of undeveloped state park. This study utilizes long term molecular marker monitoring at multiple sites in the Topanga Creek watershed to identify sources of fecal pollution and the relationship between upper and lower watershed sources. Consistent decrease in indicator bacteria and source markers downstream through the creek sites and increased bacteria levels and presence of human, gull and dog-associated markers at lagoon sites suggest an independent source near the lagoon and eliminated the creek as the source of FIB exceedances at Topanga State Beach. Dog, gull and human-associated markers were found to be important sources in the lagoon and ocean. Seasonal variability was seen for both markers, with highest levels occurring in winter. Microbial source tracking presented different trends in FIB and source markers and shows the importance of the application of a suite of markers over long term spatial and temporal sampling to identify a complex combination of chronic sources of contamination.

1. Introduction

Topanga State Beach, located between Santa Monica and Malibu CA, USA, was ranked the 9th, 4th, and 10th most impacted beaches in the state of California based on fecal indicating bacteria (FIB) levels in the 2005-2006, 2010-2011, and 2011-2012 seasons, respectively (Heal the Bay Annual Reports 2003-2013). The dominant source of FIB to the ocean has yet to be identified in spite of numerous completed projects within the lower watershed intended to improve water quality (Dagit et al., 2013). The Topanga watershed includes residential development in the upper watershed (population ???) and business development in the coastal region divided by a (???? Sized) undeveloped state park. Potential sources of fecal contamination to the watershed include malfunctioning septic systems, transients, dogs, gulls, horses farms, and wildlife. It is unclear from previous studies if Topanga Creek and a degraded lagoonal system are the dominate source of FIB to the surfzone, if Topanga Creek conveys upper watershed sources to the surf zone, or if some combination of the two results in the degraded water quality at Topanga State Beach.

A 21 month microbial source tracking (MST) study was initiated on the Topanga watershed (Figure 1) that measured FIB levels and also utilized culture-independent molecular markers for detection of host-associated fecal contamination. Unlike FIB, which can originate from multiple hosts, MST methods can help identify unique sources of fecal pollution through the use of host-associated primers that allow for identification of the original host of fecal pollution to environmental waters (Harwood et al., 2013; Boehm et al., 2013). MST methods are often deployed using a tiered approach. The first tier typically involves identifying locations impaired for FIB or other general water quality parameters (Noble et al., 2006; Boehm et al., 2003). Locations of high FIB are analyzed using spatial and temporal sampling. After which, sites impaired by FIB are also analyzed for host markers to identify sources contributing to elevated concentrations of FIB. However, results from studies using a tiered approach with FIB as a first tier to locate human-associated pollution have been mixed (Sercu et al. 2009, Reischer et al. 2008, Boehm et al. 2003, Bower et al. 2005, Flood et al. 2011). Use of FIB to infer locations for follow up analysis with source markers is confounded by the fact that FIB has been known to vary on short timescales (Boehm et al., 2002). Further, seasonal trends of host-associated markers have not been well documented and warrant further research.

This study investigates sources of FIB to the watershed and reports on the applicability of using MST technology over longer time scales. Long-term monitoring during wet and dry weather seasons, and the use of a suite of markers at many sites was utilized to characterize sources in the Topanga watershed. The sites were spaced along the main stem of the creek to determine if contamination is conveyed downstream from the town to the ocean. Impacted sites and sources and their seasonal patterns are identified to provide suggestions for targeted and effective remediation efforts to reduce number of exceedances and improve water quality at Topanga State Beach.

2. Materials and Methods

2.1 Field Site

2.1.1 Topanga Creek Watershed

Topanga State Beach receives over 750,000 annual visitors (ref). Due to the Mediterranean climate, this region experiences a dry season (April – October) and wet season (November – March), with typical rainfall averaging 20 inches a year (ref). However, rainfall during the course of this study was below average levels (9.9 inches of rainfall in 2012 – 2013)(ref). Topanga Creek watershed (approximately 47 km^2) is 70% undeveloped (GeoPentech, 2006) and includes a creek and dynamic lagoon system (Fig. 1) that breaches and berms throughout the year, contributing variable flow to coastal waters (ref). The lagoon system has been reduced from a historical size of approximately 30 acres to 2 acres by coastal development (ref).

2.1.2 Sampling sites

Based on previous studies (ref RCDSMM studies and UCLA annual report), 10 sampling locations were chosen for long-term monitoring over a 21 month period between October 2011 and 1 June 2013. Samples were collected twice monthly at five creek sites, three lagoon sites, and two marine sites. The creek sites were located 6500 m, 4800 m, 3600 m, 1700 m, and 300 m upstream from the creek discharge point. Marine sites were sampled in the surf zone at the western edge of the outlet of the lagoon and at a site approximately 100 m west (upcoast) of the lagoon (Figure 1, Table 1).

2.2 Sample Collection

All marine and lagoon samples were collected before sunrise and creek samples were taken before exposure to direct sunlight to reduce the impact of photoinactivation on samples (Boehm 2009). Sample bottles (polypropylene plastic) were washed with 10% HCl and rinsed three times with source water before use. Two liter surface samples were collected from the creek in actively flowing sections approximately 5 inches below the surface. Samples were collected from the lagoon utilizing either a pole sampler or a bridge sampler, and from the ocean on incoming waves in knee deep surf. All samples were stored on ice within 15 minutes of collection and transported back to the lab for analysis. All lab processing was completed before a maximum holding time of 6 hours.

2.3 Rain Events

Four sampling events occurred during active rainfall over the course of the study; 10/5/11, 11/17/12, 1/24/13, and 3/8/13. Two first two events were categorized as first flush (FF) which is defined as the first storm of the season to reach at least ¾ inches in rainfall. Annual rainfall for

2013 was only half as much as the typical average for that region. A total of 35 samples were collected during active rainfall.

2.7 FIB Enumeration

Samples were processed for FIB within 6 hours of collection. Three types of FIB, Total Coliform (TC), *Escherichia coli* (EC), and *Enterococcus* (ENT) were measured with Colilert-18^{TM} and Enterolert^{TM} (IDEXX, Westbrook ME) reagents and protocols to determine the most probable number (MPN) of cells per 100 mL sample. 10 mL of sample water was diluted in 90 mL Milli-Q water containing IDEXX Colilert-18^{TM} or Enterolert^{TM} reagents, sealed in a Quanti-Tray/2000 and incubated at 35^{o}C for 18 hours (for TC and EC) or 41^{o}C for 24 hours (for ENT).

2.8 DNA Extraction

Two hundred mL of sample water was filtered through 0.4 µm polycarbonate filters (EMD Millipore, Billerica MA) in triplicate. These filters were folded inwards and transferred into individual 2 ml screw cap tubes (Sarstedt Inc., Newton NC) preloaded with acid washed glass beads (Sigma-Aldrich, St. Louis MO) and archived at -80°C until extracted. DNA extraction of the filters was conducted with the DNA-EZ ST1 Extraction Kit (GeneRite, North Brunswick NJ) following the manufacturer's protocol. Eluted DNA samples were stored at -20°C until analysis of molecular host-associated markers with qPCR.

2.9 Quantitative PCR

Four host-associated markers were deployed to identify sources of fecal pollution present within the watershed. The human-associated *Bacteroidales* HF183 TaqMan assay (Haugland et al. 2010) was used to detect presence of human fecal contamination, and the BacHum TaqMan assay (Shanks et al. 2009) was used as a second human-associated marker to confirm results. A *Catellicoccus* gull-associated Gull2 TaqMan assay (Lu et al. 2008) and the dog-associated

Bacteroidales DogBact TaqMan (Dick et al. 2005) markers were used to detect presence of non-human sources. Quantitative PCR reactions were run in triplicate with 2 μl of extracted sample DNA as a template and averaged concentrations were reported in copies / 100 mL. For qPCR, samples were scored as detected when an amplification signal greater than a fluorescence threshold of 0.03 (ΔRn) was detected within 40 thermal cycles. A qPCR maximum cycle number of 40 was determined as optimal (Boehm et al., 2013). Samples were defined as falling in the range of quantification (ROQ) if at least two of three replicates amplified with concentrations at or greater than the lowest reliably detectable standard concentration. Samples were classified as below limit of quantification (BLOQ) if two or more replicates amplified, but concentrations were below the lowest reliably detectable standard concentration. Non-detects were assigned to samples if one or less replicates were positive (ND). Quantifiable sample Cqs were converted to concentration values using a master standard curve based calibration equation.

2.10 Water chemistry/Geochemical Parameters

For marine waters, measurements of water temperature and conductivity were collected in-situ (Orion conductivity meter). Ambient water chemistry measurements including pH, dissolved oxygen (mg/L and % sat), specific conductivity (μS/cm), water temperature (ºC), air temperature (ºC), and salinity (ppt) (YSI 55 DO meter, Oakton pH meter, Oakton conductivity meter, and refractometer) were. Analysis of turbidity (NTU) was tested with the LaMotte Turbidimeter 2020 and nutrients (nitrate, nitrite, ammonia and orthophosphates (ppm)) were tested using the LaMotte Smart3 Colorimeter for a subset of samples. Flow (ft/s) was measured in creek sites using a Marsh-McBirney Flowmate 3000. In-situ water quality testing equipment was calibrated before each sampling event. The membranes and solution in the DO meters were checked and replaced as needed.

2.11 Analyses

2.11.1 Historical FIB

TC, EC, ENT and discharge values were received from the Los Angeles Department of Public Health (LADPH) for the time period between January 2005 – November 2011 (1809 sampling events, approximately 6 days per week, Monday through Saturday). LADPH notes the outflow at Topanga Lagoon and any note of flow was scored as connected. Any note of “ponded” was scored as bermed. TC and EC values recorded as >13000 were evaluated as 15000 and values recorded as <67 were evaluated as 34. For ENT values recorded as >2000 were evaluated as 3000 and values recorded as <10 were evaluated as 5.

2.11.2 Time series FIB and Markers

Before statistical treatment any sample below the limit of detection was assigned a value of half the LOD. Samples greater than the upper ROQ were set to double the maximum possible value (i.e. if the max was 24916 MPN/100 mL then the value was set to 49832).

2.11.3 Box plots (Box-and-Whisker Plot)

Box plots were generated using the default settings of R Statistical Software version 12.1 (Team 2011) and RStudio™ Integrated Development Environment version 0.97.248 (RStudio, Inc., Boston, MA, USA). Horizontal lines in the boxes represent the second quartile (median). The upper and lower edges of the boxes (hinges) extend to the 1st and 3rd quartiles. The whiskers (upper/lower adjacent) extend to the most extreme data point which is no more than 1.5 times the length of the box away from the box. Data points beyond the whiskers are declared outliers and represented with open circles.

3. Results

3.1 Historical FIB trends at marine site at creek outlet

Six years of FIB and flow data taken at Topanga State Beach provided by the Los Angeles County Department of Public Health (LADPH) was grouped by season and whether the Topanga Lagoon was closed from the ocean by a sand berm. TC, EC, and ENT concentrations were significantly higher when flow from the Topanga watershed was discharging to the ocean. Less variation is observed between Wet and Recreation Seasons (Figure 2A, B, C).

3.2 Rain effects

Creek sites, lagoon sites, and marine sites were each grouped and FIB and marker levels for these groups were compared during active rainfall versus non-rain samples (Figure 3). For the creek, lagoon, and ocean all FIB levels are elevated when raining (Figure 3A, B, C). Human (Figure 3D, E) and dog markers (Figure 3F, G) are likely elevated during rain events in the creek and lagoon but the marine group is inconclusive. Gull markers show little response to rain events (Figure 3G).

3.4 Seasonal effects

The seasonal differences in FIB and marker levels over the watershed were compared for the creek, lagoon and marine sites (Figure 4). Since this study aimed to understand long term watershed dynamics, the samples collected during active rainfall were not included in the seasonal analyses. A variety of responses to the season were observed. TC and ENT wet season levels (Figure 4A, C) in the creek are likely lower than the dry season while EC wet season levels (Figure 4B) are likely elevated at the lagoon and marine sites. There are no conclusive seasonal trends in marker levels for the creek sites (Figure 4D, E, F, G). At the lagoon and marine sites, gull maker levels (Figure 4F) are likely elevated during the wet season and dog marker levels (Figure 4G) are significantly elevated in the wet season.

3.5 Creek FIB and marker spatial trends

To investigate the spatial dynamics of FIB and marker levels in the creek, we compared creek FIB and markers on a site-by-site basis (Figure 5). For creek FIB levels, the highest values were observed at 6500m and then these levels dropped at 4800m and remained below the 6500m levels for the remainder of the creek sites (Figure 5A, B, C). Human and dog markers were the highest in the creek at 6500m but then not detected at 4800m. At site 3800m human and dog markers are detected again approximately 4 times and then only one or twice at site 1800m. Human and dog markers may have been detected 4 times at site 300m although the HF183 and BacHum results do not strongly correlated at this site (Figure 5D, E, F). Gull was detected at most once at any creek site excepting the 300m site where it was detected three times (Figure 5G).

3.6 Lagoon FIB and marker spatial trends

To investigate the spatial dynamics of FIB and marker levels in the lagoon, we compared lagoon FIB and markers on a site-by-site basis (Figure 6). No spatial FIB or human marker trends are observed across the three lagoon sites (Figure 6A, B, C, D). If the lagoon sites are grouped together then the median TC, EC, and ENT levels are 4360 MPN/100ml, 712 MPN/100ml, and 210 MPN/100ml, respectively. Human markers were sporadically detected at all lagoon sites with the highest measurements reaching 1000s of copies/100ml (Figure 6D, E). Dog and gull markers are consistently detected at high levels throughout the lagoon (dog median = 960 copies/100ml, gull median 21,480 copies/100ml). There is likely an increase in both dog and gull levels towards the lagoon outlet.

3.7 Marine FIB and marker spatial trends

To investigate the spatial dynamics of FIB and marker levels at the beach, we compared marine FIB and markers on a site-by-site basis (Figure 7). TC and ENT levels likely increase from the upcoast site (-100 m) to the site at the lagoon outlet (0 m), with TC increasing from 20 to 100 MPN/100ml and ENT from 10 to 30 MPN/100 ml. Human markers were detected 2 times at the upcoast site and 4 times at the outlet site at levels on the order of 1000 copies/100ml (Figure 7D, E). Dog and gull markers are consistently high at both sites with combined medians of 1350 and 842 copies/100ml, respectively (Figure 7F, G). Gull marker is possibly twice the level at the outlet site as the upcoast site (Figure 7G).

3.8 Time series analyses

In order to understand how the creek impacts the lagoon, which subsequently impacts the ocean, three representative sites, (Creek 300m, Lagoon 25m, and Marine 0m) were examined for FIB and source markers in a time series plot. Creek FIB levels trended upward over the dry season, peaking around both Novembers with lowest levels observed in both Februaries. From January 2012 to October 2012, TC values increased by 130 times, EC increased from non-detectable to 660 MPN/100ml and ENT increased 45 times. Similar trends are not observed with any markers (Figure 8A). Human and dog marker detections were all measured in the 2012/2013 wet season and may be associated with rain events. (Figure 8B, C). Gull was rarely detected at this site. However, the one sample that was strongly positive for the Gull marker on April 2012 (Figure 8D) corresponds with exceedances of EC and ENT (Figure 8A)

The lagoon site FIB exhibit a similar pattern as the creek site, with increases in the dry season followed by decreases in the wet, but the pattern is shifted to higher FIB values (Figure 9). Human markers do not exhibit a discernible pattern and are not detected during the summer season (Figure 9B). Dog marker increases from non-detectable levels in the 2012 dry season to a maximum level of 70,000 copies/100ml in January 2013. This increase occurs predominately over 2.5 months starting in November 2012 and declines to non-detectable levels in approximately the same amount of time after reaching the January 2013 maximum. The sampling frequency during the 2011/2012 wet season is too low to confirm the dog marker pattern observed during the 2012/2013 wet season, although the isolated sampling event in January 2012 did have the highest dog marker level observed for the 2011/2012 wet season (Figure 9C). Gull marker is consistently measured in the lagoon at high levels ranging from 10,000 to 109,000 copies/100ml over this study. The seasonal pattern is not clear but in a general sense the marker level increased over the summer of 2012 and winter of 2013 until an order of magnitude decline over the month of April 2013 (Figure 9D).

The marine site does not exhibit a clear FIB pattern (**Error! Reference source not found.**10A). Sporadic human marker detections occur exclusively during the winter seasons. Three consecutive detections in November 2012 at approximately 1000 copies/100ml do not correspond with any significant human marker detections at the lagoon site. Dog marker at the marine site exhibited a similar pattern to the lagoon site with levels increasing from 200 copies/100ml in November 2012 to 42,000 copies/100ml by February 2013 and then declining to non-detectable levels by May 2013 (Figure 10C). Gull marker at the marine site also showed a similar pattern as the lagoon site with an increase over the summer into the wet season with a possible decline over April 2013 (Figure 10D). The median gull marker level at the marine site is 17 times lower than the median level at the lagoon site (Figure 10D, 9D).

4. Discussion

http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1115625/

4.1 Principal findings

1) Topanga Creek/Lagoon is a major source to surf zone

2) Rainfall washes in FIB / human / dog

3) Seasons have big (marker specific) impact on watershed (need table).

2) Sources of human and dog found in upper watershed. – around town and easy access

2) Decoupling of the upper watershed from lower watershed – remote site (ST) clean

3) Time series indicates -- human sporatic, gull endemic, dog is winter problem

Long-term sampling in the Topanga watershed has allowed for a broad look at weather, seasonal, and spatial trends to show any major patterns within the watershed. Active rainfall in the watershed increased all FIB and marker levels except the Gull marker.

When just Topanga Creek was considered, site Owl Falls (6500m OF), located just downstream of all upper watershed inputs, was found to be the most impacted, with high levels of TC, Winter EC, ENT, human markers and Winter Dog marker. These high levels were not propagated down creek as evidenced by the site, Scratchy Trail (4800m ST) having the lowest FIB and markers levels within the sampled creek sites . Sources contributing fecal contamination within the upper watershed were independent from the lower watershed sources. Therefore, effective mitigation efforts aimed to improve water quality at Topanga State Beach should focus on lower watershed sources.

If just the lagoon was considered, the PCH Bridge (HB), Topanga Lagoon (TL) and Lagoon Outlet (LO) sites were found to be consistent both by site and season, with the exception of Dog marker which was almost 100 times higher in the Winter season. Lagoon FIB and Gull marker values were higher in magnitude than creek and ocean sites. If just the ocean was considered, then FIB values were consistent by season, but may be slightly higher at site Beach Outlet (BO) than Beach Upcoast (BU). All marker values were higher in the winter season than the dry season, with Dog marker increasing by as much as 100 times. Additionally Dog marker could have been higher at site BU than BO (Figure 5F).

Long term microbial source tracking, over a 21 month period, allowed for analysis of seasonal fluctuations in molecular markers. Both Gull and Dog-associated markers were present at high frequency and were identified as important sources to Topanga Lagoon and State Beach. Gulls and other waterfowl have been found to impair water quality at other beaches and may be responsible for exceedances of FIB in surfzone and lagoon samples (Lu et al., 2008; Sinigalliano 2013). Although the gull marker was detected consistently in the lagoon, it is possible that increased number of beach visitors at the BU and BO prevented gulls from roosting on the sand; therefore a reduction in gull is observed at ocean sites in the dry season. Lafferty and colleagues (2013) found reduced shorebird populations present in beaches with increased human activity and off-leash dogs. Some gulls exhibit migrating behaviors, and it has been documented that larger shorebird populations are present in Southern California beaches in winter months (Lafferty 2001; Hubbard and Dugan 2003). This study found presence of gull waste most frequently corresponding to typical peaks in shorebird abundance (Oct – Dec months). Greater frequency of detection and magnitude of the Dog-associated marker in Winter months may also be related to the lifeguard presence at Topanga State Beach. Decreased lifeguard hours in winter months correspond to a peak of this marker in the winter season. Higher Dog marker levels in BU over BO could be due to the fact that the lifeguard patrol ended just east (downcoast) of BU, and many of the residences along the beach upcoast have dogs. Although it is difficult to control for fecal contributions from wildlife, fecal waste from pets can be mitigated by watershed managers as current regulations prevent dogs on this beach. Increased enforcement by lifeguards, community education and awareness, along with better signage may help reduce contamination associated with domestic dogs.

Human-associated markers were detected periodically in lagoon samples. Presence of markers corresponded with recorded visual observations of human feces and transient activity (Table 4). A mass balance of one direct deposit (~200g of human feces) was calculated to result in an exceedance of ENT in the lagoon. Homeless encampments were found and dismantled throughout the watershed, this a continuing issue for the city. Transient activity near the lagoon was recoded for several months (January to March 2013) adjacent to the lagoon (HB and TL sites) and found to directly impact water quality near these sites.

4.2 Strengths and weaknesses of the study

Strengths = historical monitoring, length of this study, multiple markers

Weaknesses = unclear implications of marker levels (decay rate, etc.), no strong correlation btwn markers and FIB levels. Confusing and unpredictable lagoon outflow. Need better modeling of ocean currents (not just lifeguard reports).

4.3 Strengths and weaknesses in relation to other studies

(discussing particularly any differences in results)

4.4 Meaning of the study

(possible mechanisms and implications for clinicians or policymakers)

Long-term microbial source tracking at several locations in the Topanga Creek watershed allowed for identification of several problem areas requiring remediation efforts. While human sources are prevalent near the town of Topanga. Additionally, a reduction in FIB and source markers downstream implied a decoupling of sources in creek sites and downstream lagoon and ocean sites. Dog and Gull markers presented seasonal trends, with higher levels in Winter months. Human marker was detected in lagoon and ocean samples, coinciding with presence of transient activity and two leaking septic systems. Mitigation efforts to reduce exceedances of FIB at Topanga State Beach should prioritize potential sources from the lower watershed. Testing and repairs of local sewage infrastructure, along with better enforcement regarding presence of dogs on the beach may help to improve water quality. This study showed the need for long-term water quality monitoring efforts with multiple host markers when trying to identify sources of fecal contamination.

4.5 Unanswered questions and future research

Testing of the septic systems along Pacific Coast Highway found that the system at the Ranger residence located in the State Park, was possibly leaking so repairs were completed in Summer 2013. The system at the Feed Bin was also found to be a potential source of leakage and required repair and further testing to evaluate the input of potential contamination into Topanga Creek. The other systems within Topanga State Park did not appear to be leaking, nor did the County Lifeguard facility. Although testing in Summer 2013 indicated that the majority of septic systems from businesses adjacent to Topanga Lagoon were not likely to be actively contributing any leakage during this study period, there have been several studies that indicate long lag time between input into the ground water table and emergence in either the ocean or a lagoon (Stone Environmental 2004). Therefore, human fecal contamination detected at the lagoon could possibly be partially due to two leaking septic systems (Ranger residence and Malibu Feed Bin), tested in this study. Since most of these systems have only been capped since 2008, additional dye testing in the future may be required in order to conclusively document any potential inputs. Additionally, the Los Angeles County Lifeguard Station restroom facility at Topanga Beach was upgraded in 2008 with a state of the art Advantex treatment system (Dagit et al., 2013). The renovated system incorporated chlorination, de-chlorination, and UV treatment to eliminate bacterial contamination; consistently low to non-detectable levels of FIB sampled from the lifeguard station eliminated this OWTS as a source of human pollution to the beach. Detection of the human marker from treated septage (1,785 – 893,000 copies/mL) was expected as the human-associated markers have been measured from treated wastewater effluent at similar levels in other studies (Bae and Wuertz 2009).

5. Conclusions

Acknowledgements

Numerous Resource Conservation District of the Santa Monica Mountains and University of California at Los Angeles staff, students and volunteers helped with sample collection and processing. Further assistance and support was provided by the rangers and staff at Topanga

State Park, as well as the lifeguards at Topanga Beach, and the staff of Los Angeles County Department of Beaches and Harbors. Funding was provided by Los Angeles County and by the California State Water Resources Control Board Clean Beach Initiative Grant through the Source Identification Protocol Project.

6. References

Boehm, A.B., Grant, S.B., Kim, J.H., Mowbray, S.L., McGee, C.D., Clark, C.D., Foley, D.M. and Wellman, D.E., 2002. Decadal and shorter period variability of surf zone water quality at Huntington Beach, California. Environmental Science & Technology 36(18), 3885-3892.

Boehm, J. Fuhrman, R. Mrse, and S. Grant., 2003. Tiered approach for identication of a human fecal pollution source at a recreational beach: case study at Avalon Bay, Catalina Island, California. Environmental Science &Technology, (37):673 – 680.

Boehm AB, et al., 2009. Covariation and photoinactivation of traditional and novel indicator organisms and human viruses at a sewage-impacted marine beach. Environ. Sci. Technol. 43:8046–8052.

Boehm, A. B., L. Van De Werfhorst, J. Griffith, P. Holden, J.A. Jay, O. Shanks, D. Wang and S. Weisberg., 2013 Performance of forthey-three microbial source tracking methods: A twenty-seven lab evaluation study. Water Research. In Press.

Bower, P.A., Scopel, C.O., Jensen, E.T., Depas, M.M. and McLellan, S.L. (2005) Detection of genetic markers of fecal indicator bacteria in Lake Michigan and determination of their relationship to Escherichia coli densities using standard microbiological methods. Applied and Environmental Microbiology 71(12), 8305-8313.

Dagit, R., S. Williams and J. Fuhrman. 2004. Topanga Creek Watershed Water Quality Study Final Report. Resource Conservation District of the Santa Monica Mountains, Topanga, CA.

Dagit, R., Krug, J., Adamek, K., Riedel, T., Zimmer-Faust, A.G., Thulsiraj, V., Jay, J.A., Braband, S., Tufto, D., Sherman, R. 2013. Topanga Source ID Study Annual Report 2012 - 2013 Report. Resource Conservation District of the Santa Monica Mountains, Topanga, CA.

Dick L.K., Bernhard A.E., Brodeur T.J., Santo Domingo J.W., Simpson J.M., Walters S.P., Field K.G., 2005. Host distributions of uncultivated fecal *Bacteroidales* bacteria reveal genetic markers for fecal source identification. Appl Environ Microbiol. 71(6):3184-91.

Dugan J.E., Hubbard, D.M., McCrary, M.D., Pierson, M.O., 2003. The response of macrofauna communities and shorebirds to macrophyte wrack subsidies on exposed sandy beaches of southern California. Estuarine Coastal Shelf Sci 58:25-40.

Flood C., Ufnar, J., Wang, S., Johnson, J., Carr, M. and Ellender, R. (2011) Lack of correlation between enterococcal counts and the presence of human specifc fecal markers in Mississippi creek and coastal waters. Water Research 45, 872-878.

GeoPentech, 2006. Hydrogeologic Study, Lower Topanga Creek Watershed, Los Angeles County. California. Resource Conservation District of the Santa Monica Mountains.

Harwood, V.J., Staley, C., Badgley, B.D., Borges, K., Korajkic, A., 2013. Microbial source tracking markers for detection of fecal contamination in environmental waters: relationships between pathogens and human health outcomes. FEMS Microbiology Reviews. In press.

Haugland, R.A., M. Varma, M. Sivaganesan, C. Kelty, L. Peed, O.C. Shanks. 2010. Evaluation of Genetic Markers from the 16S rRNA gene V2 Region for Use in Quantitative Detection of Selected *Bacteroidales* species and Human Fecal Waster by Real Time PCR. Syst Appl Microbiol. 2010 Oct;33(6):348-57.

Heal the Bay. 2013. 2012-2013 Annual Beach Report Card. Heal the Bay, Santa Monica, CA pg 79.

Ishii, S., Hansen, D.L., Hicks, R.E. and Sadowsky, M.J. (2007) Beach sand and sediments are temporal sinks and sources of Escherichia coli in lake superior. Environmental Science & Technology 41(7), 2203-2209.

Lafferty K.D., 2001. Birds at a Southern California beach: seasonality, habitat use and disturbance by human activity. Biodivers Conserv 10(11):1949-1962.

Lu J, Santo Domingo JW, Lamendella R, Edge T, Hill S. (2008) Phylogenetic diversity and molecular detection of bacteria in gull feces. Appl Environ Microbiol. Jul;74(13):3969-76.

Noble, R.T., Moore, D.F., Leecaster, M.K., McGee, C.D. and Welsberg, S.B. 2003. Comparison of total coliform, fecal coliform, and enterococcus bacterial indicator response for ocean recreational water quality testing. Water Research 37(7), 1637-1643.

Noble, R.T., Griffith, J.F., Blackwood, A.D., Fuhrman, J.A., Gregory, J.B., Hernandez, X., Liang, X.L., Bera, A.A. and Schiff, K., 2006. Multitiered approach using quantitative PCR to track sources of fecal pollution affecting Santa Monica Bay, California. Applied and Environmental Microbiology 72(2), 1604-1612.

R Development Core Team (2011). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org/.

Reischer, G.H., Haider, J.M., Sommer, R., Stadler, H., Keiblinger, K.M., Hornek, R., Zerobin, W., Mach, R.L. and Farnleitner, A. (2008) Quantitative microbial faecal source tracking with sampling guided by hydrological catchment dynamics. Environmental Microbiology 10(10), 2598-2608.

Sercu, B., VanDeWerfhorst, L., Murray, J. and Holden, P. (2009) Storm Drains are Sources of Human Fecal pollution During Dry Weather in Three Urban Southern California watersheds. Environmental Science and Technology 43 (2), 293-298.

Shanks, O.C., C.A. Kelty, M. Sivaganesan, M. Varma, and R.A. Haugland. 2009. Quantitative PCR for Genetic Markers of Human Fecal Pollution. Applied and Environmental Microbiology 75:5507-5513.

Sinigalliano, C.D., Ervin, J.S., Van De Werfhorst, L.C., Wang, D., Wanless, D., Bartkowiak, J., Layton, B., Raith, M., Schriewer, A., Badgley, B.D., Lee, C., Goodwin, K.D., Lee, J., Boehm, A.B., Noble, R.T., Holden, P.A., Jay, J.A., Wuertz, S., Byappanahalli, M.N., Whitman, R.L., Sadowsky, M.J., Meijer, W., Balleste, E., Gourmelon, M., Griffith, J.F., Ryu, H., Domingo, J.W.S., 2013. Multi-laboratory assessment on the performance of PCR assays targeting Catellicoccus marimammalium for microbial source tracking of coastal birds. Water Research In Press.

Stone Environmental. 2004. Risk Assessment of Decentralized Wastewater Treatment Systems in High Priority Areas in the City of Malibu, California. Final Report August 30, 2004. Prepared for the Santa Monica Bay Restoration Commission by Stone Environmental Inc. Montpelier, VT.

Surbeck, C.Q., Jiang, S.C., Ahn, J.H. and Grant, S.B. (2006) Flow fingerprinting fecal pollution and suspended solids in stormwater runoff from an urban coastal watershed. Environmental Science & Technology 40(14), 4435-4441.

Yamahara, K.M., Walters, S.P. and Boehm, A.B. (2009) Growth of Enterococci in Unaltered, Unseeded Beach Sands Subjected to Tidal Wetting. Applied and Environmental Microbiology 75(6), 1517-1524.

Tables:

Table 1: Location of sample sites.

Site Type	Site Distance from outlet m	Easting m	Northing m	Elevation m
Marine	-100	353726	3767515	0
Marine	0	353896	3767506	0
Lagoon	1	353872	3767529	0
Lagoon	25	353887	3767573	0
Lagoon	35	353868	3767649	0
Creek	300	354015	3767841	0
Creek	1700	354075	3768713	0
Creek	3600	353522	3770391	200
Creek	4800	353518	3771500	500
Creek	6500	352673	3772373	700

Table 2: Molecular marker qPCR assays and associated calibration values from master standard curves.

Name	Target	Forward Primer / Reverse Primer	Probe/Dye	Amplicon Size (bp)	Reference	Calibration
HF183 Taqman	Human associated *Bacteroides* 16S	ATCATGAGTTCACATGTCCG / CGTAGGAGTTTGGACCGTGT	FAM-CTGAGAGGAAGGTCCCCCACATTGGA-TAMRA	167	Haugland et al., 2010	
BacHum	Human associated					
DogBac	Dog associated					
Gull2 Taqman	Gull associated *Catellicoccus marimammalium*	TGCATCGACCTAAAGTTTTGAG/ GTCAAAGAGCGAGCAGTTACTA	FAM-CTGAGAGGGTGATCGGCCACATTGGGACT-BHQ1	412	Shibata et al., 2010	

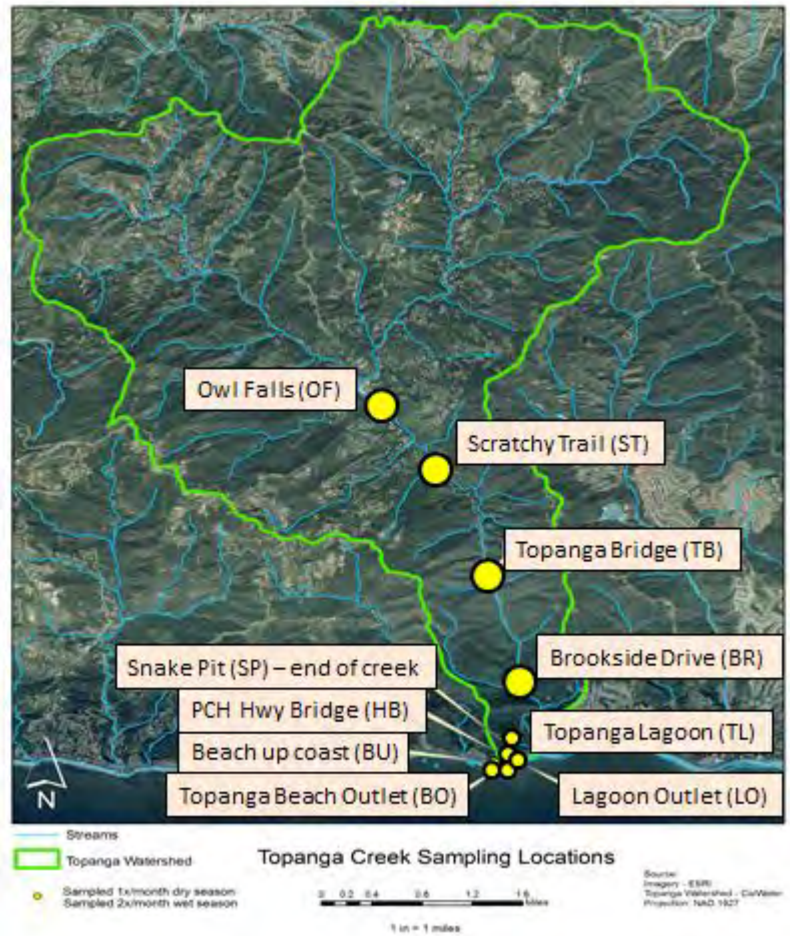

Figure 1. Map of the Topanga Creek Watershed and sampling locations. Five samples were collected from the lower half of the watershed along the main stem of Topanga Creek. Creek sites include Owl Falls (OF), Scratchy Trail (ST), Topanga Bridge (TB), Brookside Drive (BR) and Snake Pit (SP). Three samples were collected within Topanga Lagoon at the Pacific Coast Highway Bridge (HB), Topanga Lagoon (TL) at the east end, and at the Lagoon outlet (LO), just before the lagoon discharge point. Two marine samples were taken, one directly out from the lagoon (BO), this sample represents the mixing point if the lagoon is breached, and an upcoast beach site (BU) north of the lagoon.

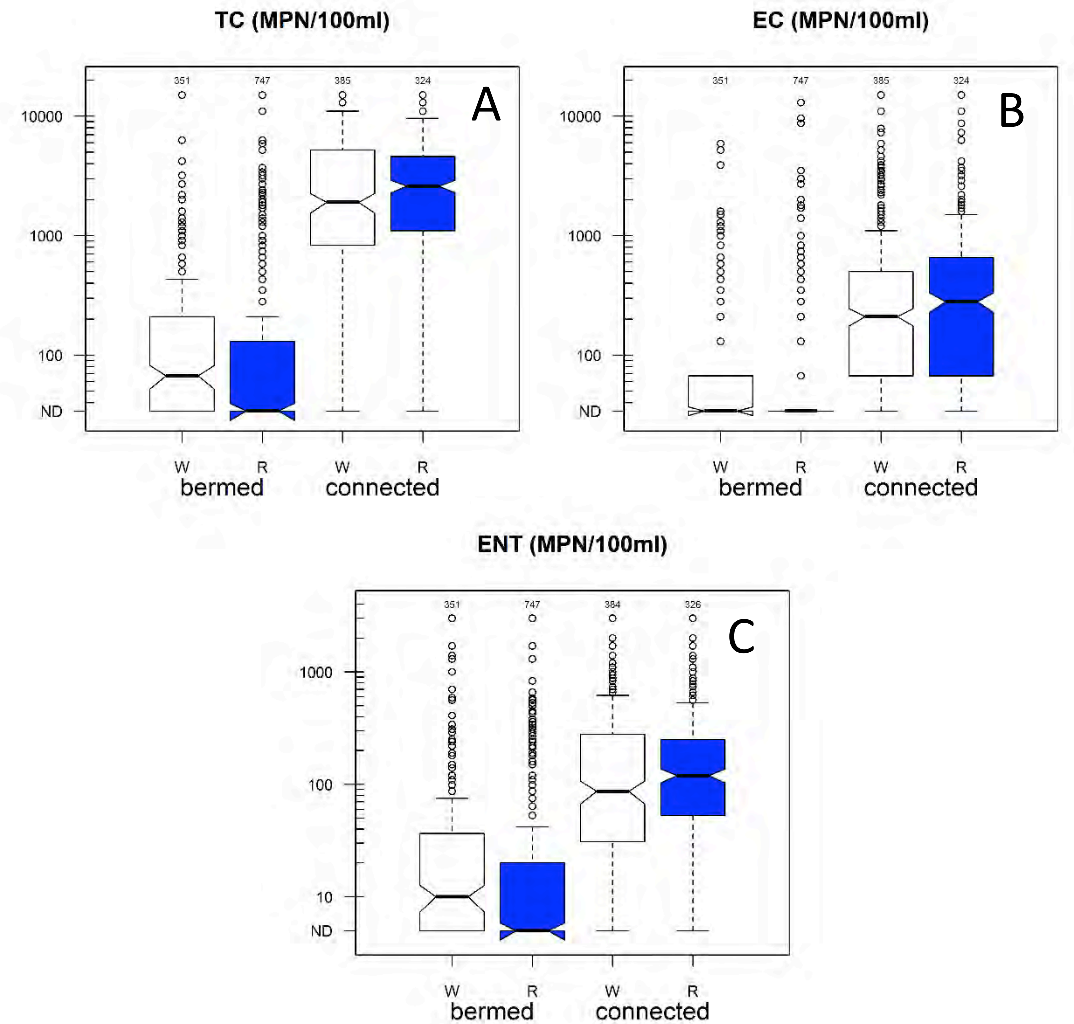

Figure 2: Lagoon outflow and seasonal effects on historical FIB levels Topanga Beach. Box-and-Whisker plots of A) TC, B) EC, and C) ENT compared by winter season (W), recreation season (R), if the creek was flowing (connected), or if the creek was not flowing (bermed). Quantities along top of plot indicate number of samples per group.

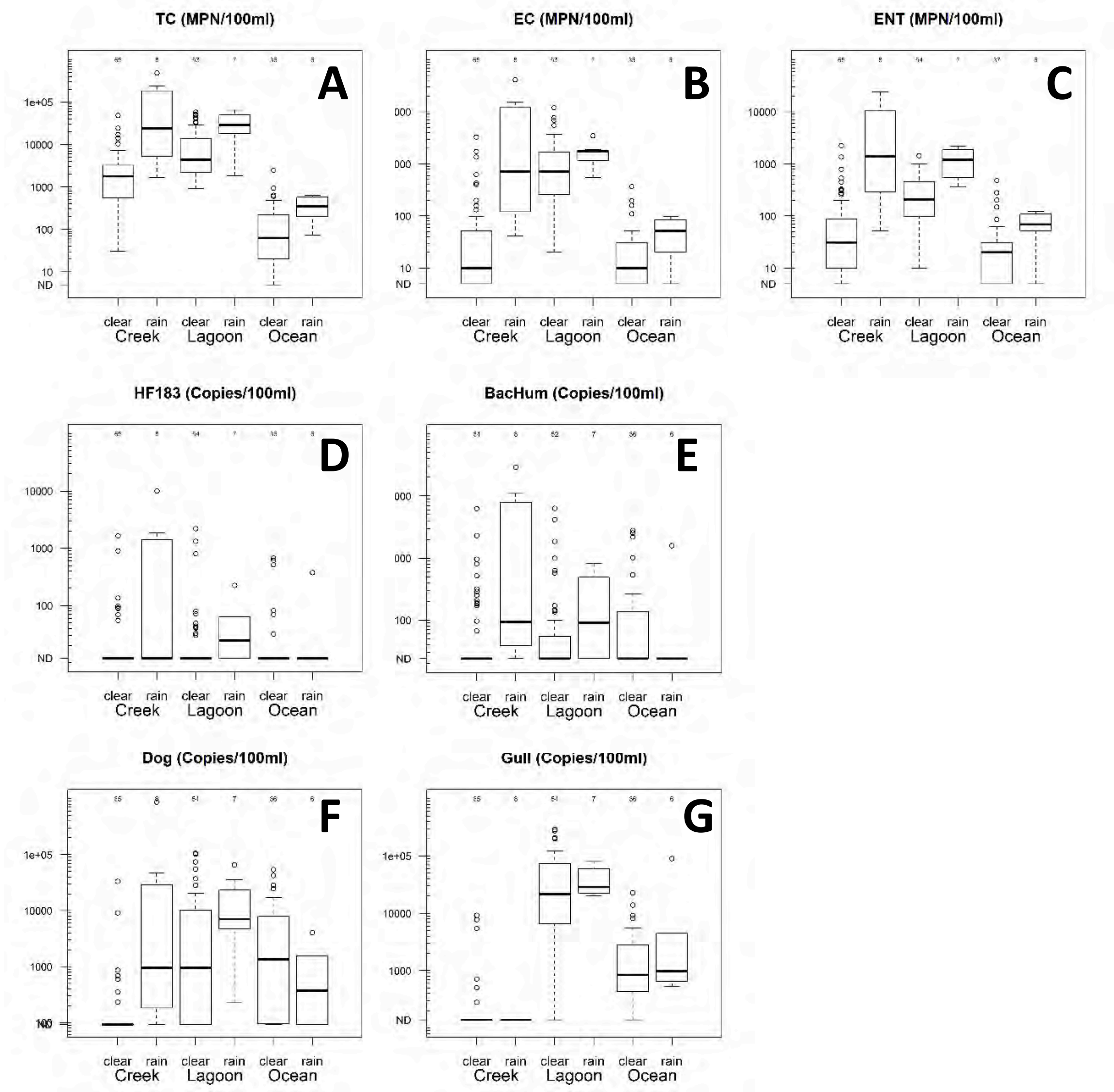

Figure 3: Effect of active rainfall during sampling on FIB and marker levels at Topanga Creek, Lagoon and Beach. Box-and-Whisker plots of A) TC, B) EC, C) ENT, D) HF183, E) BacHum, F) DogBac and G) Gull2 values recorded during active rainfall (rain) versus non-raining (clear) conditions. Quantities along top of plot indicate number of samples per group.

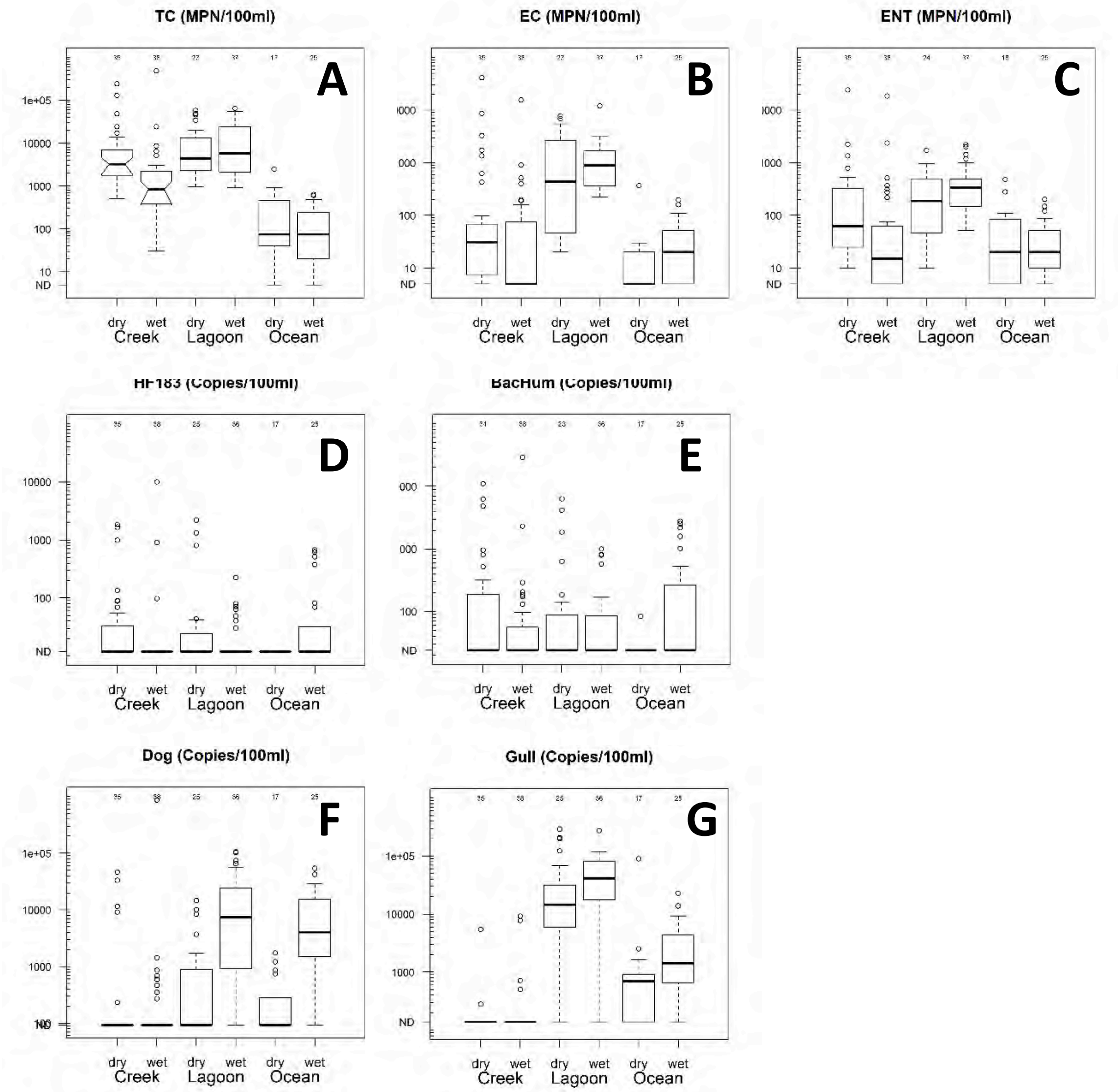

Figure 4: Seasonal effects on FIB and marker levels on Topanga Creek, Lagoon and Beach. Box-and-Whisker plots compare recreational season (dry) versus wet season (wet) of A) TC, B) EC, C) ENT, D) HF183, E) BacHum, F) DogBac and G) Gull2 values. Quantities along top of plot indicate number of samples per group. Samples collected during active rainfall excluded from analysis.

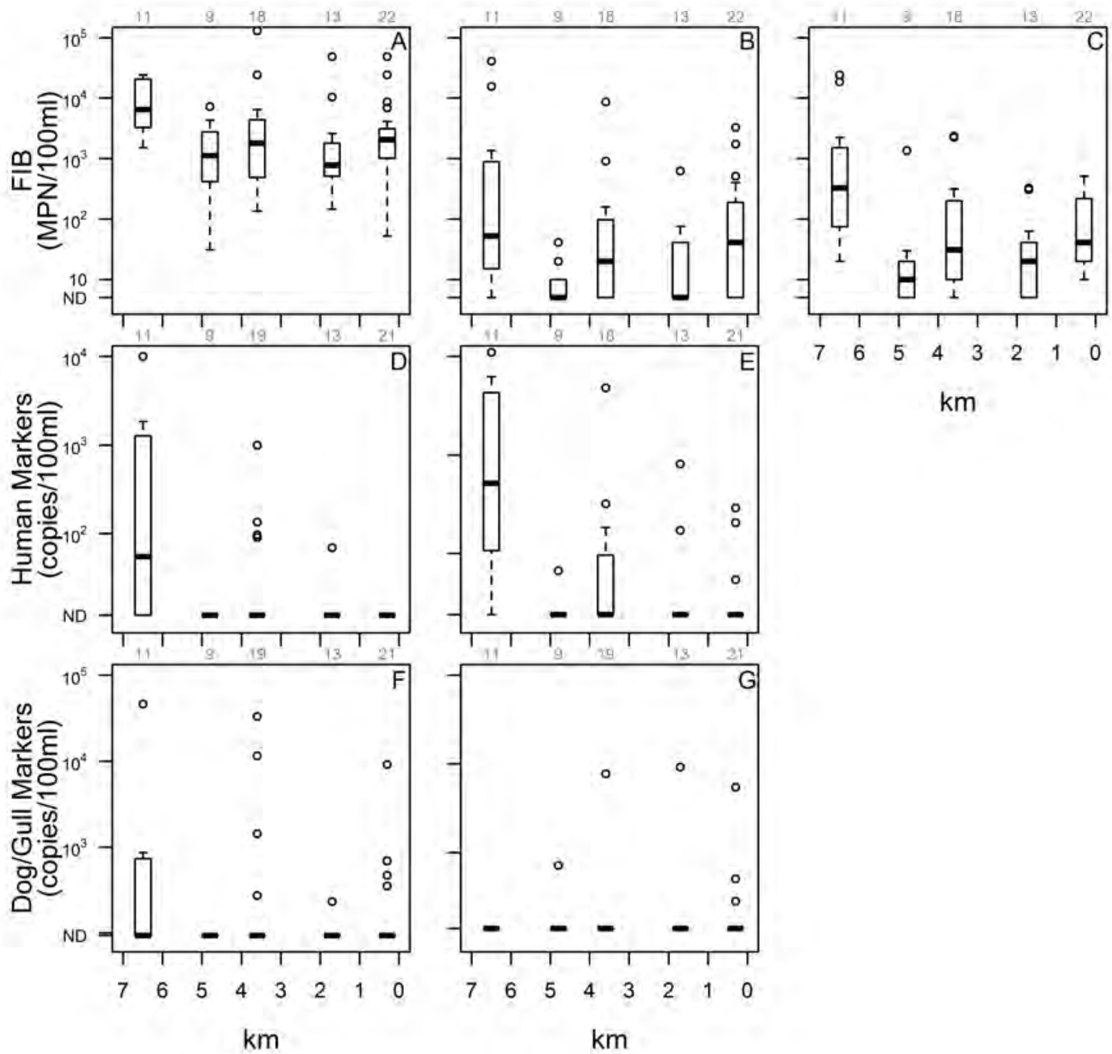

Figure 5. FIB and marker levels at Topanga Creek sites. Box-and-Whisker plots of FIB levels A) TC, B) EC, C) ENT, and human marker levels D) HF183 and E) BacHum, and F) dog and G) gull levels for each site. The km value indicates the distance up-creek from the sample site to the terminus of the creek. Quantities along top of plot indicate number of samples per group. Samples collected during active rainfall excluded from analysis.

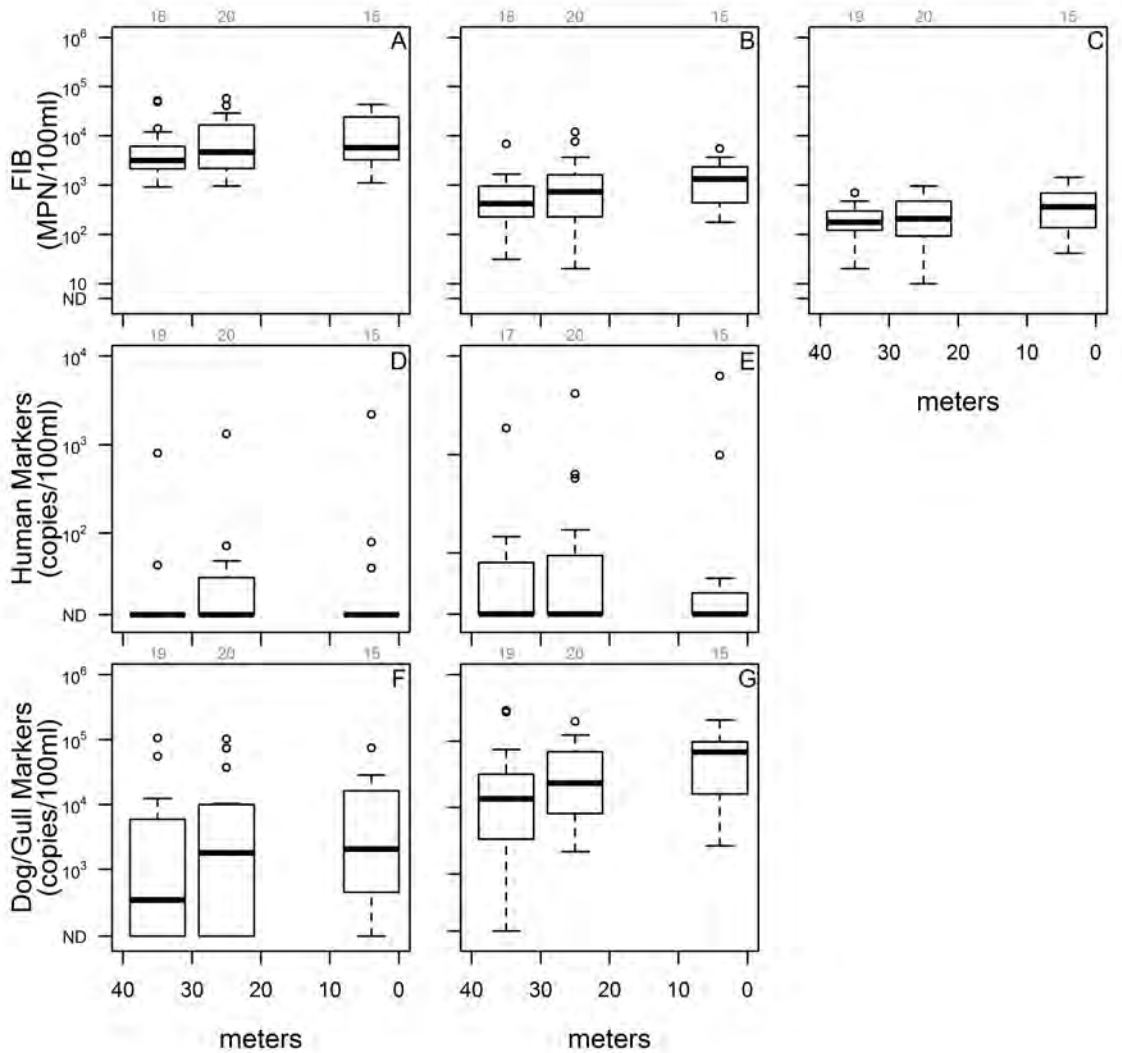

Figure 6. FIB and marker levels at Topanga Lagoon sites. Box-and-Whisker plots of FIB levels A) TC, B) EC, C) ENT, and human marker levels D) HF183 and E) BacHum, and F) dog and G) gull levels for each site. The meter value indicates the distance up-creek from the sample site to the terminus of the creek. Quantities along top of plot indicate number of samples per group. Samples collected during active rainfall excluded from analysis.

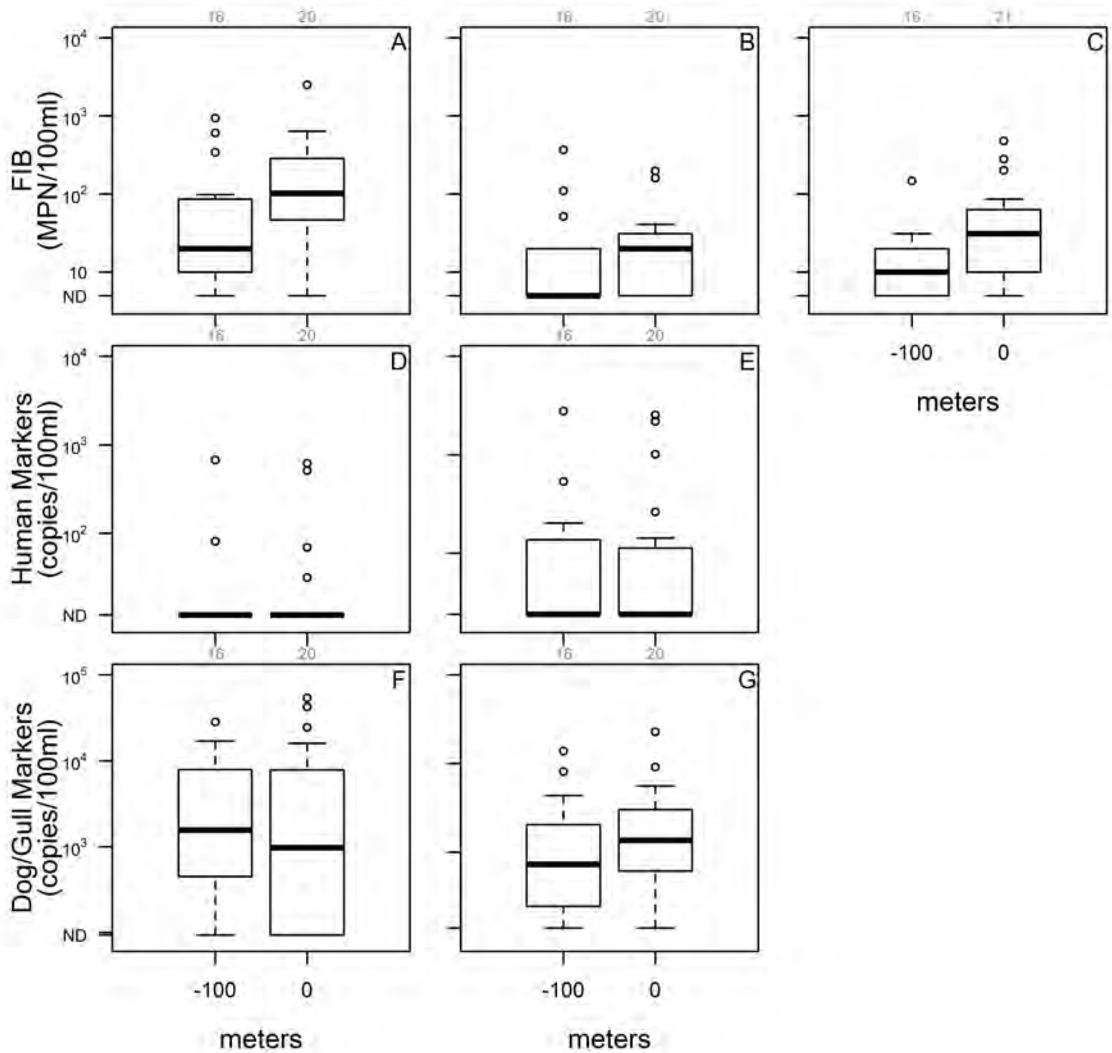

Figure 7. FIB and marker levels at Topanga Marine sites. Box-and-Whisker plots of FIB levels A) TC, B) EC, C) ENT, and human marker levels D) HF183 and E) BacHum, and F) dog and G) gull levels for each site. The meter value indicates the distance up-coast (eastward) from the sample site to the terminus of the creek. Quantities along top of plot indicate number of samples per group. Samples collected during active rainfall excluded from analysis.

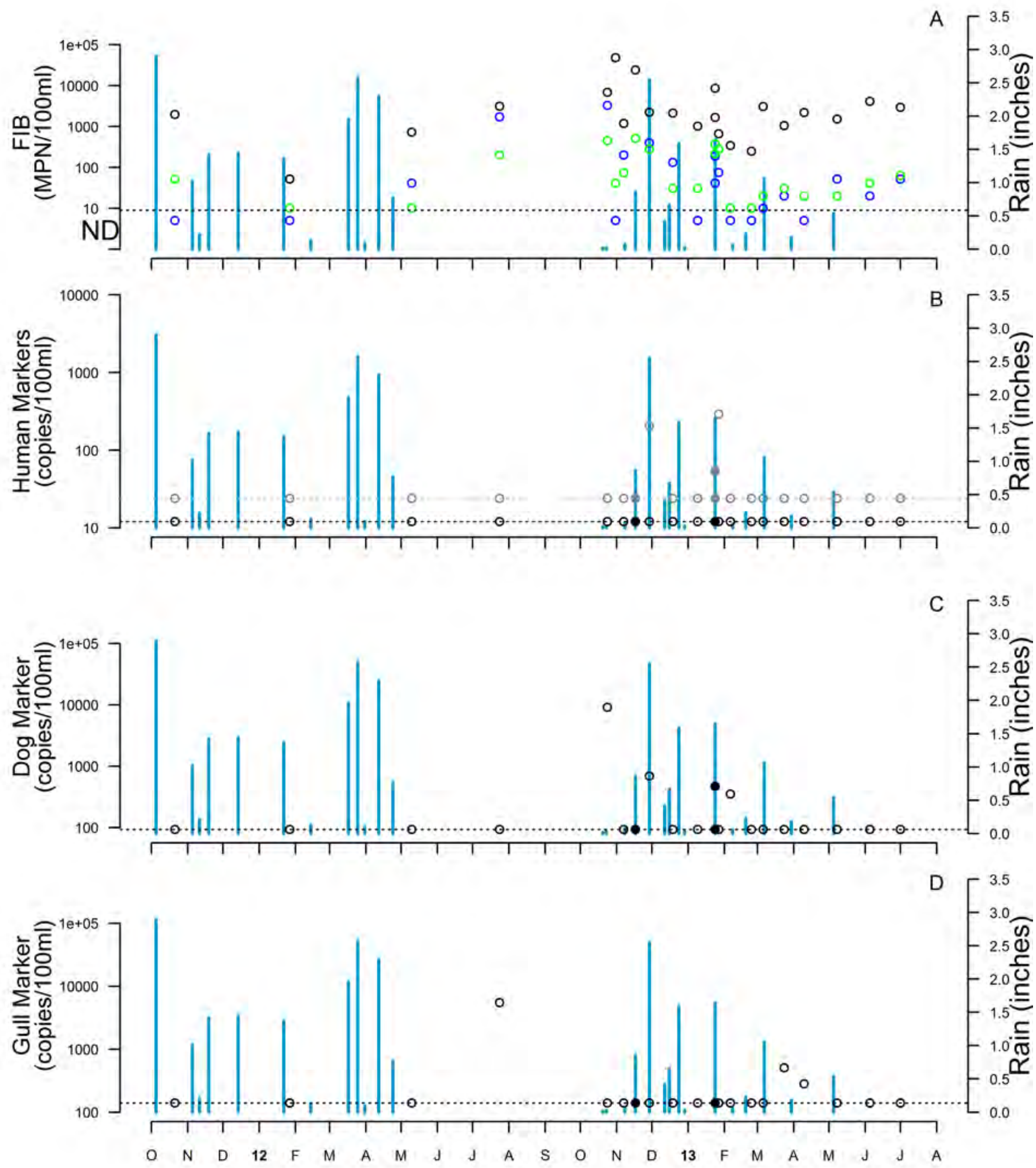

Figure 8. Time-series plots for creek site at 0.3 km showing trends in A) TC (black circles), EC (blue circles), and ENT (green circles), B) human (HF183 black circles, and BacHum grey circles), C) dog and D) gull markers. Dotted lines represent the limit of detection. 3 day running average rainfall level indicated by the blue bars.

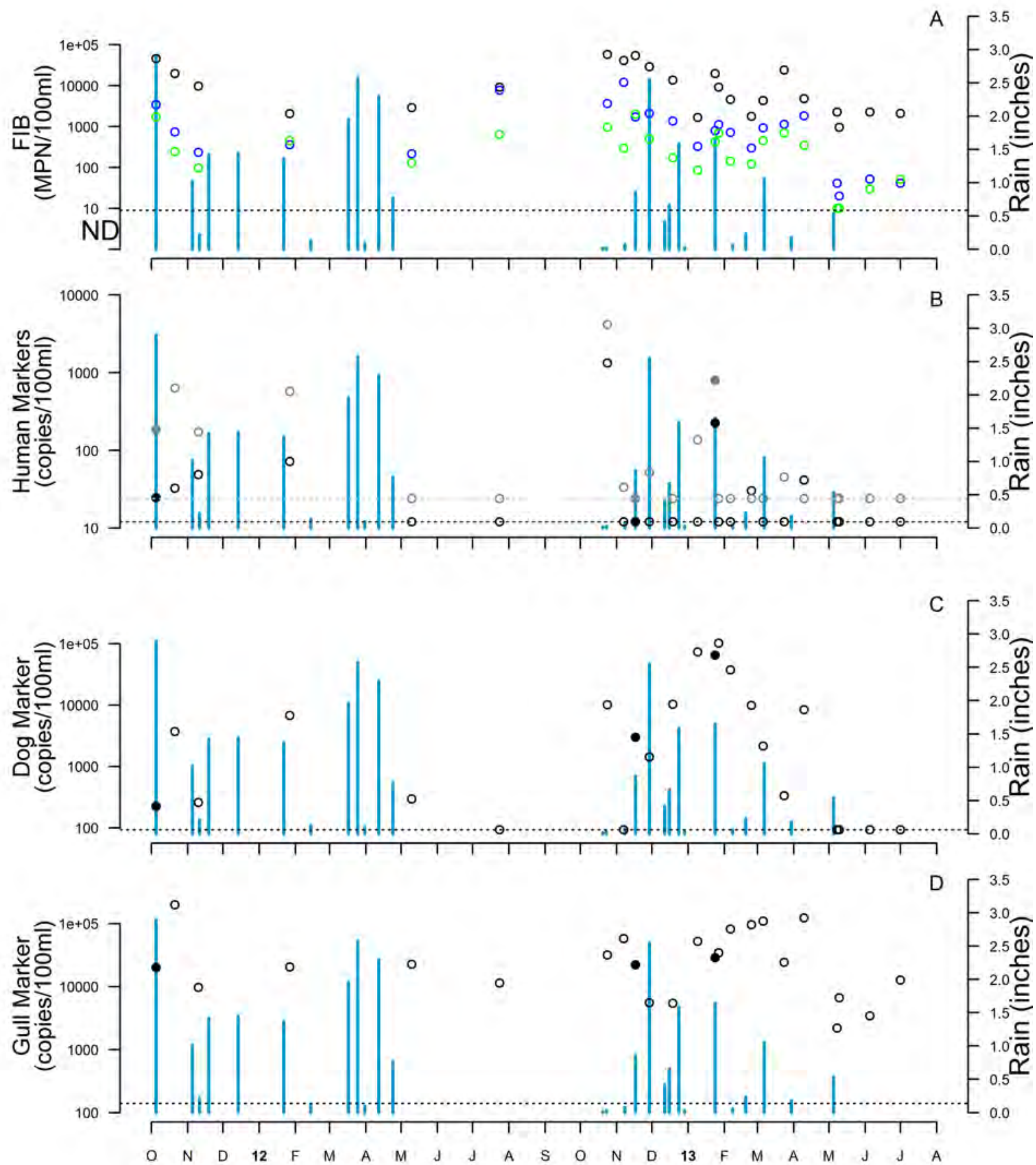

Figure 9. Time-series plots for lagoon site (25 m) showing trends in A) TC (black circles), *E. coli* (blue circles), and ENT (green circles), B) human (HF183 black circles, and BacHum grey circles) and C) dog and D) gull-associated markers. Dotted lines represent the limit of detection. 3 day running average rainfall level indicated by the blue bars.

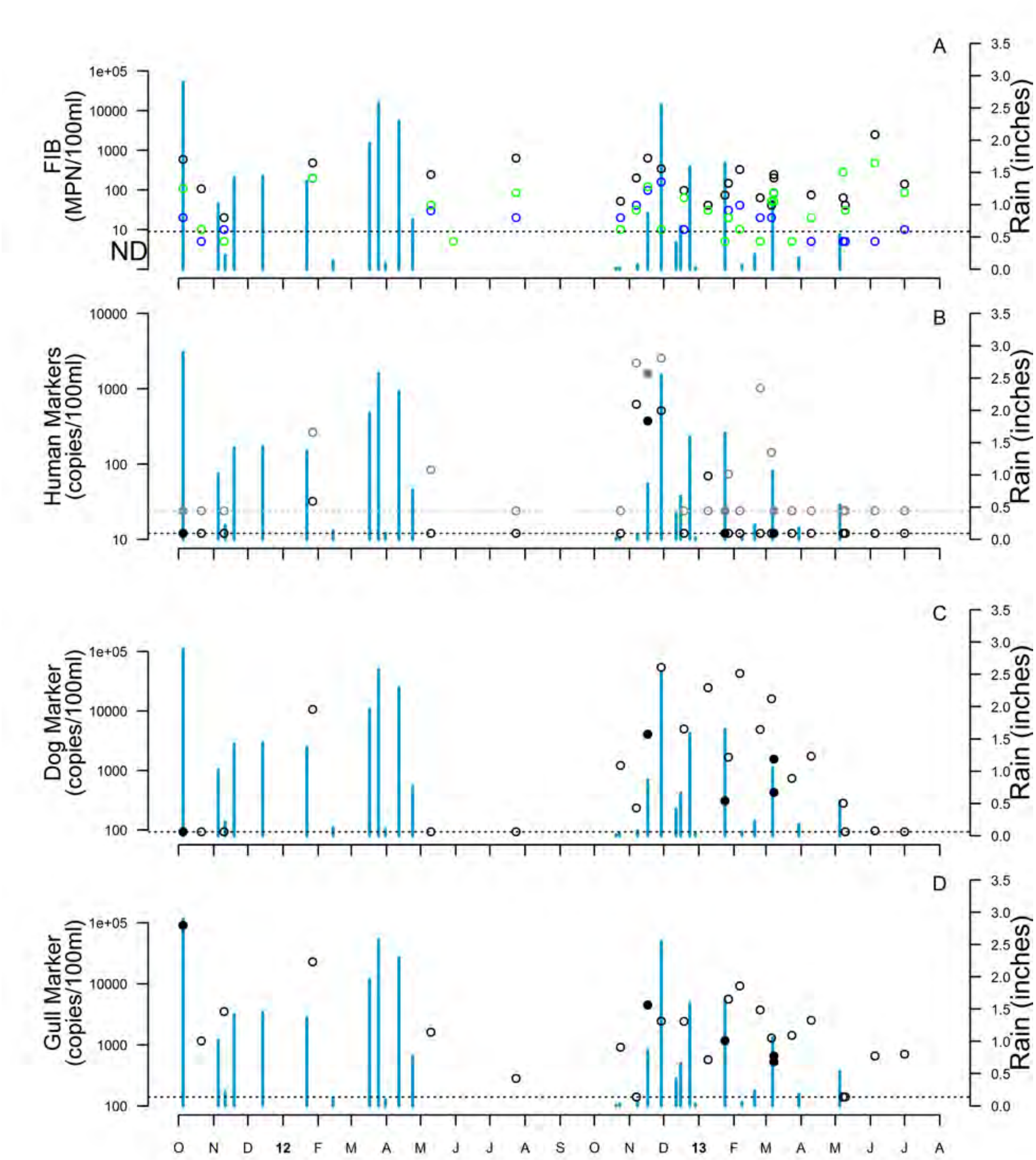

Figure 10. Time-series plots for marine site at creek terminus showing trends in A) TC (black circles), *E. coli* (blue circles), and ENT (green circles), B) human (HF183 black circles, and BacHum grey circles) and C) dog and D) gull-associated markers. Dotted lines represent the limit of detection. 3 day running average rainfall level indicated by the blue bars.