Contents lists available at ScienceDirect



Journal of Hydrology: Regional Studies

journal homepage: www.elsevier.com/locate/ejrh



Identifying locations of sewage pollution within a Hawaiian watershed for coastal water quality management actions

Check for updates

Tracy N. Wiegner^{a,*}, Steven L. Colbert^a, Leilani M. Abaya^{a,1}, Jazmine Panelo^a, Kristina Remple^b, Craig E. Nelson^b

^a Marine Science Department, University of Hawai'i at Hilo, 200 W. Kawili St., Hilo 96720, HI, USA ^b Daniel K. Inouye Center for Microbial Oceanography Research and Education, Department of Oceanography and Sea Grant College Program, University of Hawai'i at Mānoa, 1950 East West Road, Honolulu 96822, HI, USA

ARTICLE INFO

Keywords: Sewage OSDS FIB Stable nitrogen isotopes Macroalgae Dye tracer studies

ABSTRACT

Study region: Puakō, Hawai'i Island.

Study focus: Locations of sewage pollution in the Puakō watershed were identified through measurements of sewage indicators at groundwater wells and within Puakō's and adjacent resorts' shoreline waters. Dye tracer tests, water quality, $\delta^{15}N$ macroalgal, and $\delta^{15}N$ - and $\delta^{18}O$ -NO₃⁻ measurements, along with stable isotope mixing models, were combined to assess water quality impairment caused by different Onsite Sewage Disposal System (OSDS) types, and used to predict water quality improvements from future management actions.

New hydrological insights for the region: Sewage indicators were highest within Puakō's shoreline waters, including: Enterococcus spp., Clostridium perfringens, human-associated Bacteroides, and δ^{15} N-NO₃⁻. Mixing model results using δ^{15} N- and δ^{18} O-NO₃⁻ suggest that sewage was a dominant NO₃⁻ source, comprising > 40% at 10 of the 16 shoreline stations. δ^{15} N macroalgae measurements confirmed presence of sewage at most stations. In groundwater wells and at adjacent resorts' shoreline waters, sewage indicators were low, and δ^{15} NO₃⁻ was indicative of soils and fertilizers. Puakō dye tracer tests revealed that sewage reached the shoreline within 5 h to 10 d, and that OSDS type did not affect travel time. Water quality was similar in front of homes with different OSDS. In conclusion, sewage is entering the groundwater at Puakō, and the underlying geology, rather than OSDS type, primarily controls the speed at which sewage reaches the shoreline. Our findings highlight the need for improved sewage treatment and collection at Puakō.

* Corresponding author.

E-mail addresses: wiegner@hawaii.edu (T.N. Wiegner), colberts@hawaii.edu (S.L. Colbert), l.abaya@hawaiianspringswater.com, abaya@hawaii.edu (L.M. Abaya), jrpanelo@hawaii.edu (J. Panelo), remple@hawaii.edu (K. Remple), craig.nelson@hawaii.edu (C.E. Nelson).

¹ Current address: Hawaiian Springs, LLC., 16–305 Old Volcano Road. Kea'au HI 96749.

https://doi.org/10.1016/j.ejrh.2021.100947

Received 24 May 2021; Received in revised form 6 October 2021; Accepted 10 October 2021

Available online 19 October 2021

Abbreviations: OSDS, Onsite Sewage Disposal Systems; STP, Sewage Treatment Plant; ATU, Aerobic Treatment Unit; HDOH, Hawai'i Department of Health; FIB, Fecal Indicator Bacteria; MST, Microbial Source Tracking; SGD, Submarine Groundwater Discharge; HBP, Hapuna Beach Prince; MKB, Mauna Kea Beach; FO, Fairmont Orchid; ML, Mauna Lani; HEW, high- elevation wells; MEW, mid-elevation wells; LEW, low-elevation wells; DL, detection limit; TDN, total dissolved nitrogen; TDP, total dissolved phosphorus; IRMS, isotope ratio mass spectrometer; USGS, U.S. Geological Survey; ANOVA, analysis of variance; SIAR, stable isotope analysis in R; MPN, most probable number; CFU, colony forming units.

^{2214-5818/© 2021} The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).



Fig. 1. Sixteen stations (stars south to north: 1–16) along the Puakō coastline, Hawai'i Island, and two resort stations (black triangles) north (17,18) and south (19,20) of Puakō were chosen to evaluate fecal indicator bacteria, nutrients, δ^{15} N- and δ^{18} O-NO₃, and δ^{15} N in macroalgal tissue. Shoreline stations in Puakō were delineated into three regions: south (stations 1–6), middle (stations 7–10), and north (stations 11–16). Groundwater samples were collected at high (open squares, HEW), mid (open circles, MEW), and low (open triangles, LEW) elevations and analyzed for sewage indicators. In addition, dye tracer studies were conducted at 10 different locations to determine the hydrological connection between aerobic treatment units (yellow circles, ATUs), cesspools (green circles), and septic tanks (orange circles) to nearshore waters. The Mauna Lani sewage treatment plant (STP) is a potential location being considered tor receiving sewage from Puakō. (For interpretation of the references to colour in this figure, the reader is referred to the web version of this article.)

1. Introduction

Worldwide, coastal communities are impacted by sewage pollution. It is a complex problem because sewage is a cocktail of elevated and potentially hazardous concentrations of pathogens, nutrients, cleaning chemicals, hydrocarbons, and pharmaceuticals (Wear and Vega Thurber, 2015). It poses human health risks to recreational water users, as well as causes environmental degradation. Human health impacts include abdominal, skin, urinary, and blood infections (Pinto, 1999). Annually, there are over 120 million gastroenteritis cases worldwide associated with sewage contaminated waters (Shuval, 2003). Additionally, one of the most sensitive ecosystems to sewage pollution is coral reefs (Wear and Vega Thurber, 2015), and they are one of the most economically valuable and biologically diverse ecosystems on Earth (Knowlton et al., 2010). In 2020 U.S. dollars, coral reefs are globally worth \$35.8 billion (Conservation International, 2008). Elevated nutrient concentrations from sewage pollution can stimulate benthic macroalgae resulting in phase shifts from coral- to macroalgal-dominated reefs (Hunter and Evans, 1995; Lapointe et al., 2005). Nutrients also affect coral growth rates, species distributions and abundance, and coral community diversity (Pastorok and Bilyard, 1985; Parsons et al., 2008). Sewage pollution has been linked to coral disease prevalence and severity, as well as decreased calcification rates and increased bioerosion (Sutherland et al., 2010; Redding et al., 2013; Yoshioka et al., 2016; Prouty et al. 2017). Sewage impacts to both human and coral reef health are costly; globally, the annual economic burden of recreational waterborne illnesses is \$16.4 billion (2018 U.S. dollars; Shuval, 2003) and \$32.9 billion for loss in ecosystem services of coral reefs (2018 U.S. dollars; Cesar et al., 2003).

Sewage enters waterbodies from sewage treatment plant (STP) outfalls and injection wells, as well as from onsite sewage disposal systems (OSDS) like cesspools, septic tanks, and aerobic treatment units (ATUs). OSDS are commonly used in rural areas, and are more widely used in Hawai'i than any other state in the country (HDOH, 2015). Hawai'i Department of Health (HDOH) estimates that there are presently 110,000 OSDS statewide, with 49,000 classified as cesspools on Hawai'i Island (Whittier and El-Kadi, 2014). In 2015, HDOH banned construction of new cesspools due to concerns about threats to human and coral reef health; it was the last state to do so following Rhode Island, who banned them in 1968 (HDOH, 2015). In 2017, Hawai'i State passed legislation to replace all cesspools by 2050 (HDOH, 2017). With sea level rise, it is anticipated that the frequency and magnitude of sewage pollution water quality issues will increase as more OSDS become inundated (Elmir, 2018; McKenzie et al., 2021).

Sewage pollution is a chronic environmental problem in Hawai'i, first documented in Kāne'ohe Bay, O'ahu, in 1951 (Smith et al., 1981), and it is an ongoing issue throughout the state today. One area of concern in Hawai'i is the South Kohala region on Hawai'i Island. This area has the largest contiguous coral reef in the state and is thought be impacted by land-based pollution, including sewage (NOAA, 2016). Puakō, a coastal community in this region, has some of the richest coral reefs in Hawai'i, but their condition has been continually declining since the 1970s; coral cover has decreased by 50%, and macroalgal cover has increased by a similar percentage over this time period (Minton et al., 2012; HDAR, 2013). It is thought in part that sewage pollution is contributing to the decline of the reef. Concerns over sewage pollution at Puakō began in the 1960s (Schott, 2010). As a result, in 1990, Puakō was designated as a Critical Wastewater Disposal Area by Hawai'i County. These are areas where the disposal of sewage has or may cause adverse effects on human health or the environment due to existing hydrogeological conditions. As a result, homeowners building new homes or renovating existing ones are required to install septic tanks. With a surface elevation that is only 1–5 m above mean sea level, OSDS and their drainage fields are in close proximity to groundwater.

Since 2014, researchers have been working to document the presence of sewage pollution along Puakō's shoreline, its transport offshore, and contact with the coral reef and possible effects on reef condition (Couch et al., 2014; Yoshioka et al., 2016; Abaya et al., 2018a, 2018b; Aguiar, 2020). As part of this ongoing effort, Puakō community members wanted to know if sewage pollution in their nearshore waters was from their homes, or from adjacent resort developments, or upslope residential communities. This information is critical for management decisions being considered by the Puakō community for reducing sewage pollution in their nearshore waters. Alternatives to cesspools and septic tanks under consideration are: converting all cesspools to ATUs, or connecting to an existing nearby STP or building one within their community (Aqua Engineering, 2015).

The goals of this study were to: 1) identify the location within the Puakō watershed where sewage was entering into the groundwater using biological and chemical sewage indicators, 2) determine the source of fecal indicator bacteria (FIB) and nitrate (NO_3) through microbial source tracking (MST) and stable isotopes of nitrate using mixing models, respectively, 3) quantify water quality impairment caused by homes with OSDS through dye tracer studies and measurements of sewage indicators, and 4) assess whether proposed sewage treatment upgrades are sufficient for meeting state water quality standards. This information is critical for Puakō to help guide their decision regarding upgrading sewage collection, treatment, and disposal. Approaches and results from this project provide a roadmap for future projects dealing with sewage pollution in nearshore waters throughout Hawai'i State, and other coastal communities worldwide.

2. Methods

2.1. Site description

Puakō is a 3.5-km residential, coastal community located within the South Kohala district of Hawai'i Island that is built on a 3360-y old basalt lava flow (Fig. 1; Wolfe and Morris, 1996). There has been little soil development in this arid environment, with an average rainfall range from 250 to 750 mm (Giambelluca et al., 2013). The fractured substrate is highly permeable, and rainfall primarily becomes groundwater (Engott, 2011). Across the study area is an unconfined basal aquifer that exists in horizontally extensive lava flows from Mauna Kea and Mauna Loa volcanoes (Mink and Lau, 1993). Average submarine groundwater discharge (SGD) at the shoreline ranges from 2083 to 2730 L m⁻¹ h⁻¹ (Paytan et al., 2006). Puakō has 261 lots, with 231 developed. One-hundred and

thirty-eight have OSDS – 49 are cesspools, 77 are septic tanks with leach fields, and 12 are ATUs (Aqua Engineering, 2015). There are also 70 homes where the type of OSDS is unknown (Aqua Engineering, 2015). Resorts north of Puakō, Hapuna Beach Prince (HBP, now Weston) and the Mauna Kea Beach Resorts (MKB), are hooked up to a single wastewater reclamation facility, where reclaimed water is used to irrigate the Mauna Kea Golf Course (Mauna Kea Properties Inc., 1985; Fukunaga and Assoc. Inc., 2010). The resorts south of Puakō, KaMilo at Mauna Lani (ML), the Fairmont Orchid (FO), and Fairways at Mauna Lani are connected to the Kalahuipua'a Lagoon Facility (Mauna Lani STP Inc., 2006; Aqua Engineering, 2015). Chlorinated effluent from the aerated lagoon is used to irrigate sod on the property (Mauna Lani STP Inc., 2006; Aqua Engineering, 2015). Currently, there is one development up-slope of Puakō, Waikoloa Village, which has 6362 people (U.S. Census, 2010); most homes have OSDS (1587), with the remainder (413) connected to a STP that produces R3 effluent through biofilm reactors and dissolved air flotation (HDOH, 2017; Abaya et al., 2018a).

2.2. Station selection

To determine where within the Puakō watershed sewage was entering into the groundwater, we sampled two Puakō anchialine ponds (elevation: ~ 1 m), two Puakō wells (elevation range: 1–11 m; low elevation wells, LEW), seven mid-elevation wells (MEW; elevation range: 16–44 m) within the Mauna Lani development, and three high elevation wells (HEW; elevation range: 363–368 m) within Waikoloa Village (Fig. 1). To determine where sewage was emerging along the South Kohala coast, we sampled 16 shoreline stations in Puakō (Fig. 1; *see* Abaya et al., 2018a for more details), two north of Puakō at HBP and MKB, and two south of Puakō at ML and FO (Fig. 1). The Puakō shoreline was delineated into three regions for latter data analyses: south Puakō (stations 1–6), middle Puakō (stations 7–10), and north Puakō (stations 11–16).

2.3. Sample collection and analyses

At each station, water samples were collected in sterile, acid washed, polypropylene plastic bottles and analyzed for salinity, FIB, nutrients, and $\delta^{15}N$ – and $\delta^{18}O$ - NO₃⁻. All shoreline samples were collected during low tide to capture maximum impacts of ground-water on nearshore water quality (Peterson et al., 2009), and near sunrise to minimize photo-inhibition of FIB (Fujioka et al., 1981). Salinity was measured at the time of water collection using an YSI Pro 2030 multi-parameter probe. Additionally, at all shoreline stations, macroalgal tissue samples were collected for $\delta^{15}N$ analysis. Water and macroalgal tissue samples were collected between November 2014 through March 2017.

To determine NO₃⁻ sources to shoreline waters, potential sources were sampled, including: sewage from cesspools and septic tanks (n = 3 samples; Abaya et al., 2018a), HEW (n = 9), LEW (n = 8), ambient ocean water (n = 2), well-fertilized soil (n = 2), and soil from under Kiawe trees (*Prosopis pallida*) (n = 3). Kiawe is an introduced N₂-fixing tree found widely on the leeward coasts on the Hawai'i Island, and contributes N to soil and groundwater (Dudley et al., 2014). All soil samples were dried and then shaken overnight with reagent-grade water. N source samples were collected at several locations to assess spatial variability. Samples were analyzed for δ^{15} N – and δ^{18} O-NO₃⁻ and nutrients.

2.3.1. FIB traditional culture methods

Enterococcus spp. was analyzed using the IDEXX Laboratories Inc. Enterolert method, following the manufacture's recommendations of a 1:10 dilution with sterile water (ASTM D6503–19, 2019). *Clostridium perfringens* was enumerated by filtering sample water through a 0.45-µm pore size cellulose nitrate filter (WhatmanTM) and mCP medium (Acumedia, Baltimore, MD, USA) (Bisson and Cabelli, 1979).

2.3.2. FIB molecular methods

Sample water was filtered through 0.2-µm Advantec® mixed cellulose ester membrane filters and stored frozen in Qiagen bead beating tubes (Cat. No.: 12888–100-PBT). For analysis, samples were thawed at room temperature 1–2 h and homogenized in a ThermoSavant FastPrep® FP120 Cell Disrupter for 45 s at maximum speed 6.5 m/s. DNA extraction proceeded via the Qiagen DNEasy PowerSoil column genomic DNA purification kit proceeded following the manufacturer's directions.

Detection of FIB was performed using four established qPCR assays (Appendix, Table A1). 25 μ L reaction mixtures containing 10 μ L of Kapa Probe Force Master Mix (Cat. No. KK4301), 400 nmol/L of each specific primer, 200 nmol/L of each specific probe, and 1 μ L of sample or standard DNA were amplified using an Eppendorf Realplex² Mastercycler®. A 2-min hot start at 95 °C was followed by 45 cycles of 95 °C denaturing for 30 s, and 60 °C elongation for 30 s

Standard curves were constructed from serial dilutions of either genomic DNA or synthetic DNA fragments (Appendix, Table A1). For each assay, eight dilution steps ranging from 50,000 - 10 gene copies per well were performed in triplicate on every run plate. R² values ranged from 0.94 to 0.99 and threshold cycle y-intercepts (a theoretical limit of detection) for these assays ranged from 37.7 to 39.64. Threshold values were controlled manually at 500 Relative Fluorescence Units.

Note, other *Bacteroides* markers are available for non-human animals. However, for this study, we chose to solely use the humanassociated markers as we wanted to address the sewage pollution issue relative to OSDS. Additionally, this is a residential neighborhood where other *Bacteroides* sources would be minimal. While, there are some wild pigs and goats, it would take much longer for bacteria from their feces to reach the water table at Puakō as it is very dry, especially in comparison to the OSDS, where many of them are situated within the water table.

2.3.3. Nutrients and $\delta^{15}N$ - and $\delta^{18}O$ -NO₃⁻

Water samples were filtered through pre-combusted (500°C for 6 h) GF/F filters (WhatmanTM), and stored frozen until analysis at the University of Hawai'i at Hilo (UH Hilo) Analytical Laboratory. Nutrients were analyzed on a Pulse TechniconTM II autoanalyzer using standard methods (NO₃⁻ + NO₂⁻ [Detection Limit (DL) 0.07 µmol/L, USEPA 353.4], NH₄⁺ [DL 0.36 µmol/L, USGS I-2525], PO₄³⁻ [DL 0.03 µmol/L, TechniconTM Industrial Method 155–71 W], total dissolved phosphorous (TDP) [DL 0.5 µmol/L, USGS I-4650–03], H₄SiO₄ [DL 1 µmol/L, USEPA 366]), and reference materials (NIST; HACH 307–49, 153–49, 14242–32, 194–49). Total dissolved nitrogen (TDN) was analyzed by high-temperature combustion, followed by chemiluminescent detection of nitric oxide (DL 5 µmol/L, ASTM D5176, Shimadzu TOC-V, TNM-1) (Sharp et al., 2002). For NO₃⁻ isotope analysis, samples were filtered through a 0.22-µm cellulose acetate filter (WhatmanTM) and frozen until analysis. δ^{15} N- and δ^{18} O-NO₃⁻ samples were analyzed on a Thermo-FinniganTM Delta Plus isotope ratio mass spectrometer (IRMS) with data normalized to United States Geological Service (USGS) standards (USGS32, USGS34, USGS53) at Northern Arizona University Stable Isotope Laboratory. IAEA-NO3 was used as a check standard.

2.3.4. Macroalgal tissue $\delta^{15}N$

At the time of water sample collection, the most abundant macroalgae species were sampled (~5 g) at all shoreline stations and analyzed for δ^{15} N; the number of macroalgal species collected varied with station. At Puakō, *Ulva fasciata* (23 occurrences), *Cladophora spp.* (28 occurrences), and *Gelidiella acerosa* (28 occurrences) were collected, plus 10 unknown species. At the resort shoreline stations, *Ulva spp.* (9 occurrences), *Pterocladiella spp.* (9 occurrences), and turf algae (9 occurrences) were collected. Macroalgal tissues were placed on ice during transport to the laboratory, where tissues were rinsed with deionized water. Subsamples of macroalgal tissue were preserved as voucher specimens and identified to the lowest taxonomic resolution using an OlympusTM CH30 microscope (Abbott, 1999; Abbott and Huisman, 2004). The remaining portions of the samples were dried at 60 °C until a constant weight was achieved, ground and homogenized using a Wig-L-Bug grinding mill, and ~2 mg of the macroalgal tissue were folded in 4 × 6 mm tin capsules for stable isotope analysis. Because a common macroalgal species was not found at all stations, and the number of species identified at each station varied, we collected tissues from all species present and compiled them into a composite sample, where their tissues were ground together. Macroalgal tissues were analyzed for δ^{15} N using a Thermo-FinniganTM Delta V Advantage IRMS with a Conflo III interface and a CostechTM ECS 4010 Elemental Analyzer located at the UH Hilo Analytical Laboratory. Data were normalized to USGS standard NIST 1547. Isotopic signatures are expressed as standard (δ) values, in units of parts per mil (∞), and calculated as [(R_{sample} – R_{standard}) / R_{standard}] x 1000, where R = ¹⁵N/¹⁴N. δ^{15} N macroalgal tissue values were plotted relative to δ^{15} N source values to determine their N sources (Derse et al., 2007; Wiegner et al., 2016; Abaya et al., 2018a).

2.4. Dye tracer study

Dye tracer studies were conducted to determine the hydrological connection of cesspools, septic tanks, and ATUs to nearshore waters. Ten homes were identified, four with cesspools, three with septic tanks, and three with ATUs within the Puakō community (Fig. 1). A known concentration of fluorescein, a non-toxic organic dye, was administered to the OSDS over ~ 10 h. Each hour, 50 or 100 g of dye were mixed with 20 L of tap water and slowly added to the OSDS. Additional tap water was added as needed and its volume recorded to calculate initial dye concentration. Cesspool data are from an earlier study (Abaya et al., 2018a), and these data are incorporated into the water quality impairment and improvement analysis (*see* section 2.7.3).

Six shoreline stations in front of each home (60–70 m) were sampled for the presence of dye. These stations represented groundwater springs with varying salinity, and two background stations with higher salinity and no apparent freshwater input. Water samples were collected at each station before and during the dye tracer study in opaque brown high-density polyethylene bottles to prevent photo-degradation, pre-rinsed with sample water, and stored at 4 °C until analysis. During the first 12 h of the dye study, samples were collected every two h to identify any fast-flow pathways. Afterwards, two samples were collected at each station within an hour of the lowest-low tide each day for up to 14 d.

To quantify the concentration of fluorescein, samples were brought to room temperature, filtered (GF/F WhatmanTM), and analyzed using a Turner AU10 fluorometer in the dark. Fluorescein standards were made by dilution from a 100,000 ppb stock solution to 10–200 ppb. The detection limit for our analysis was 0.95 ppb (USEPA, 2011). When salinity was not measured in the field, conductivity of samples was measured in the laboratory (Orion Star) and converted to PSS-78 salinity (UNESCO, 1981).

Water and macroalgae samples were also collected in front of the homes to assess water quality conditions. Stations were determined based on where dye emerged at springs. Water samples were collected from a single station in front of each home three times. Water samples were analyzed for FIB, nutrients, and stable isotopes of NO₃⁻. Macroalgae were analyzed for δ^{15} N.

2.5. Data analyses

2.5.1. Statistical analyses

To determine if FIB, nutrients, and δ^{15} N- and δ^{16} O-NO₃⁻ values differed among watershed locations and shoreline regions in Puakō (Fig. 1), one-way analysis of variance (ANOVA) tests were used for each variable. Two-sample *t*-tests were used to compare differences among these parameters between the north and south resorts. Data were tested for normality and equal variances. If assumptions were not met for parametric analyses, log transformations were used. All statistical analyses were conducted using Minitab17 (2010) with $\alpha = 0.05$.

(1)

2.5.2. SIAR mixing models

To determine the relative percent contributions of different N sources to NO₃ pools among and within watershed locations, we used each water types' δ^{15} N- and δ^{18} O-NO₃- values, along with δ^{15} N- and δ^{18} O NO₃⁻ from N sources, and analyzed them in a mixing model (Stable Isotope Analysis in R (SIAR) v. 4.0). Note, two isotopes are required for the model to run; our primary isotope of interest was δ^{15} N. The first step in this analysis was to visualize the stable isotope data for samples relative to values for the sources. To do this, biplots of δ^{15} N- and δ^{18} O-NO₃⁻ were created, graphing averages for each station relative to averages for all potential NO₃⁻ sources (data not shown). These figures were used to decide on which sources to include in SIAR mixing models. This analysis also allowed us to determine if different sources have overlapping stable isotope values. If there is overlap, it is harder to distinguish each source's contribution to a sample from one another in the modeling effort. Based on this analysis, we decided to include sewage, groundwater, and kiawe/fertilized soil in the mixing models. Ocean water was not included as a NO₃⁻ source as its NO₃⁻ concentrations were below detection limits for stable isotope analysis (> $2 \mu mol/L$, Coplen et al., 2012), and therefore, considered to be negligible. Two mixing models were executed. The first model examined potential NO3⁻ sources to mid-elevation groundwater wells, low-elevation groundwater wells, Puako's anchialine ponds, the resorts' shoreline waters, and Puako's shoreline waters. In this model, data from all stations within a water type were averaged and station differences within water types were not examined. The second model then examined potential NO₃ sources to Puakō's and the resorts' shoreline stations. This analysis was done to identify any shoreline sewage pollution hot spots. The SIAR mixing model uses a Bayesian framework which takes into account natural variability and uncertainty within a system by allowing multiple sources of uncertainty to be incorporated (Parnell et al., 2010). The Shapiro-Wilk test was used to verify the normality of the isotope data for the three NO_3^- sources, and these data were normal. Relative percent contributions are reported as 50% Bayesian credibility intervals, allowing a wider range of variability to be reported.

2.5.3. Quantifying water quality impairment from OSDS

Dye concentration vs. time data were used to calculate flow rate and travel time of the dye between the OSDS and the ocean. To determine if FIB, nutrients, stable isotopes of NO_3^- , and $\delta^{15}N$ macroalgal tissue differed in front of homes with different OSDS types, a one-way ANOVA was used. A mixing model in SIAR was used to partition potential NO_3^- sources in front of homes with different types of OSDS. In addition, we calculated water quality improvement with regards to FIB at the shoreline that will occur by switching from the current distribution of OSDS to all ATUs or a STP.

A mixing model was used to estimate the fraction of FIB (*Enterococcus* spp. and *C. perfringens*) from sewage in groundwater sampled at shoreline springs and predict improvements in FIB concentration that would occur by upgrading cesspools. Of the three end-member sources of spring water considered (sewage, groundwater, and seawater), sewage is the dominant source of FIB, with concentrations more than two orders of magnitude greater than the other sources, based on end-member FIB concentrations measured in this and earlier studies (Abaya et al., 2018a, 2018b). Assuming sources other than sewage are negligible at contributing FIB to the spring water, we simplified the model so that FIB concentrations at the spring (C_t) are solely derived from sewage, either cesspools, septic systems, or ATUs:

$$fC_r + (1-f)eC_r = C_t$$

Table 1

Average \pm SE of *Enterococcus* spp. *Clostridium perfringens*, human-associated *Bacteroides* (markers HF183 and BacHum; 1 +log 10 scale) and δ^{15} N $-NO_{3^{-}}$ and $\delta^{18}O -NO_{3^{-}}$ in water samples for high elevation wells (Waikoloa Village), mid elevation wells (Mauna Lani), low elevation wells (Puakō wells), resort shoreline stations (Mauna Kea, Hapuna Prince, Fairmont Orchid, and Mauna Lani), Puakō shoreline stations and Puakō ponds (anchialine ponds) on Hawai'i Island. Number of stations sampled within each watershed category varied between 3 and 16 (station n) and the total number of observations per location per parameter are footnoted below. Human-associated *Bacteroides* measurements for the resorts are not available (n/a). Superscript letters indicate significant groups from One-way ANOVA and post-hoc Tukey's tests. $\alpha = 0.05$.

	1	0 0 1		1	5		
Watershed Location	Station (n)	Enterococcus spp. (MPN/100 mL)	C. Perfringens (CFU/100 mL)	HF183 (Human) (Copies/100 mL)	BacHum (Human) (Copies/100 mL)	δ ¹⁵ N –NO ₃ - (‰)	δ ¹⁸ O –NO ₃ - (‰)
High Elevation Wells	3	15 ± 9^{c}	0 ± 0^d	$\textbf{0.00} \pm \textbf{0.00}$	$\textbf{0.00} \pm \textbf{0.00}$	$\textbf{4.47} \pm \textbf{0.21}^{d}$	1.98 ± 0.43^{bc}
Mid Elevation Wells	6	5 ± 0^{c}	0 ± 0^d	$\textbf{0.00} \pm \textbf{0.00}$	$\textbf{0.00} \pm \textbf{0.00}$	6.37 ± 0.40^{bc}	1.23 ± 0.32^{c}
Low Elevation Wells	2	119 ± 67^{bc}	$0\pm 0^{a\text{-}d}$	$\textbf{0.00} \pm \textbf{0.00}$	$\textbf{0.05} \pm \textbf{0.04}$	$6.29\pm0.70^{a\text{-}d}$	$2.85\pm0.60^{a\text{-}c}$
Resort Shoreline	4	32 ± 14^{c}	$0\pm 0 ^{cd}$	n/a	n/a	$5.16\pm0.31~^{cd}$	2.61 ± 0.70^{bc}
Puakō	16	410 ± 83^{b}	4 ± 1^a	$\textbf{0.58} \pm \textbf{0.16}$	0.71 ± 0.17	8.64 ± 0.25^a	$\textbf{7.92}\pm0.02^{a}$
Puakō Ponds	2	8165 ± 2935^a	$18\pm11^{\text{a-c}}$	$\textbf{0.47} \pm \textbf{0.28}$	1.74 ± 0.55	$\textbf{8.59}\pm\textbf{0.41}^{ab}$	$\textbf{4.19} \pm \textbf{0.53}^{ab}$

Number of observations per location, per parameter:

High Elevation Wells: Enterococcus = 6, C. perfringens = 6, HF183 (Human) = 6, BacHum (Human) = 6, $\delta^{15}N - NO_{3^-} = 11$, $\delta^{18}O - NO_{3^-} = 11$ Mid Elevation Wells: Enterococcus = 16, C. perfringens = 16, HF183 (Human) = 11, BacHum (Human) = 11, $\delta^{15}N - NO_{3^-} = 25$, $\delta^{18}O - NO_{3^-} = 25$ Low Elevation Wells: Enterococcus = 6, C. perfringens = 6, HF183 (Human) = 4, BacHum (Human) = 4, $\delta^{15}N - NO_{3^-} = 7$, $\delta^{18}O - NO_{3^-} = 7$ Resort Shoreline: Enterococcus = 12, C. perfringens = 12, $\delta^{15}N - NO_{3^-} = 12$, $\delta^{18}O - NO_{3^-} = 12$

Puakō Shoreline: Enterococcus = 137, C. perfringens = 137, HF183 (Human) = 48, BacHum (Human) = 48, $\delta^{15}N - NO_{3^-} = 48$, $\delta^{16}O - NO_{3^-} = 48$ Puakō Ponds: Enterococcus = 7, C. perfringens = 5, HF183 (Human) = 4, BacHum (Human) = 4, $\delta^{15}N - NO_{3^-} = 7$, $\delta^{18}O - NO_{3^-} = 7$



V

Fig. 2. Spatial distribution of sewage indicators in the Puakō watershed, including shoreline stations at adjacent resorts north (Mauna Kea, Hapuna Prince) and south (Fairmont Orchid, Mauna Lani) of the community on Hawai'i Island. A) *Enterococcus* spp., B) *Clostridium perfringens*, C) Human-associated *Bacteroides* (HF 183 and BacHum markers), and D) δ^{15} N-NO₃⁻. Puakō watershed samples were collected from June – August 2016; resort samples were collected from January–March 2017.

In this equation, f is the fraction of cesspools in the neighborhood, C_r is the FIB concentration in raw sewage, and e is the fraction of FIB remaining after treatment by septic systems and ATUs. At Puakō, OSDS are 44% cesspools, 48% septic systems, and 8% ATUs (Aqua Engineering, 2015). Septic tanks decrease *Enterococcus* spp. and *C. perfringens* by 50% and 68%, respectively, based on published studies (Chauret et al., 1999; Withers et al., 2011). Since no published information regarding the effectiveness of ATUs removing FIB was found, we assumed the same reduction as septic systems. This assumption may underestimate FIB reduction in ATUs, as residence time and concentration of oxygen can differ between ATUs and septic tanks. Oxygen can affect the survival of FIB differentially, as *Enterococcus* spp. can survive in aerobic conditions, while *C. perfringens* is an obligate anaerobe.

By converting all cesspools to septic systems or ATUs, the new concentration at a spring would be: $C_t' = eC_r$. The percent reduction of FIB at the spring was then calculated as $R = (C_t - C_t')/C_t$. Combining these with Eq. 1, the percent reduction of FIB at the spring is simplified to:

$$R=1-[e/(f+(1-f)e)]$$
(2)

The predicted spring concentration after transition away from cesspools was calculated as $C_t'=C_t(1-R)$. In addition to assuming sewage is the only source of FIB to spring water, this model assumes that different sewage sources are well mixed before groundwater discharge at springs (e.g., hydrodynamic dispersion by tides), and that the fraction of sewage in the spring water does not change after upgrades. So, while the FIB concentration may decrease, other components of sewage, like nutrients, may or may not change, depending on how the OSDS treats that component of the sewage.

3. Results

3.1. FIB

Enterococcus spp. concentrations significantly differed among watershed locations (p < 0.0001, Table 1), but were similar among Puakō's shoreline regions. Puakō's anchialine ponds had *Enterococcus* spp. concentrations that were an order of magnitude higher (range: 324–19,018 MPN/100 mL) compared to other watershed locations (Fig. 2A, Table 1). Puakō's shoreline waters and the low-elevation wells had the second highest *Enterococcus* spp. concentrations (0–7985 MPN/100 mL). Mid- and high-elevation wells, as well as shoreline waters at the resorts, had similar concentrations, and they were the lowest of all the watershed locations sampled (0–147 MPN/100 mL, 4 out of 34 samples were above 35 MPN/100 mL). In comparison to *Enterococcus* spp., *C. perfringens* was rarely detected at the different watershed locations (Fig. 2B; Table 1). In fact, it was only detected in Puakō's anchialine ponds and shoreline waters (0–90 CFU/100 mL, 39 of 141 samples were above 5 CFU/ 100 mL) (Table 1). Like *C. perfringens*, human-associated *Bacteroides* (HF183 and BacHum markers) were only detected in Puakō's anchialine ponds and shoreline waters (Fig. 2C, Table 1).

3.2. Nutrients

Nutrient concentrations tended to be higher in inland waters (wells, anchialine ponds) and were lower at the shoreline (Table 2).

Table 2

Average \pm SE of salinity, turbidity, and nutrient concentrations (NO₃- + NO₂-, NH₄+, TDN, PO₄³⁻, H₄SiO₄) in water samples for high elevation wells (Waikoloa Village), mid elevation wells (Mauna Lani), low elevation wells (Puakō wells), resort shoreline stations (Mauna Kea, Hapuna Prince, Fairmont Orchid, and Mauna Lani), Puakō shoreline stations, and Puakō ponds (anchialine ponds) on Hawai'i Island. Number of stations sampled within each watershed category varied between 3 and 16 (station n) and the total number of observations per location per parameter are footnoted below. Superscript letters indicate significant groups from One-way ANOVA and post-hoc Tukey's tests. $\alpha = 0.05$.

		-			-			
Watershed Location	Station (n)	Salinity	Turbidity (NTU)	NO ₃ - + NO ₂ - (µmol/L)	NH ₄ + (µmol/ L)	TDN (µmol/ L)	PO4 ³⁻ (µmol/ L)	H ₄ SiO ₄ (μmol/L)
High Elevation Well	3	0.18 ± 0.01^{b}	0.39 ± 0.07^{c}	95.27 ± 2.33^{ab}	2.29 ± 0.89^{ab}	$102\pm1^{a\text{-}c}$	2.35 ± 0.06^a	775 ± 53^a
Mid Elevation Well	6	1.76 ± 0.09^{b}	0.84 ± 0.15^{bc}	120.35 ± 4.62^a	1.46 ± 0.36^{b}	$133\pm8^{\text{a}}$	1.54 ± 0.05^{ab}	760 ± 28^a
Low Elevation Well	2	1.91 ± 0.27^{b}	$\textbf{6.88} \pm \textbf{2.74}^{ab}$	114.55 ± 8.93^a	3.31 ± 1.12^{ab}	127 ± 15^{ab}	1.94 ± 0.57^{ab}	657 ± 76^a
Resorts Shoreline	4	22.37 ± 2.93^a	1.11 ± 0.18^{bc}	$\textbf{47.01} \pm \textbf{9.68}^{b}$	0.80 ± 0.18^{b}	57 ± 11^{c}	1.01 ± 0.19^{b}	328 ± 77^{b}
Puakō	16	21.53 ± 0.76^a	0.23 ± 0.02^{b}	$65.51 \pm 5.21^{\mathrm{b}}$	1.64 ± 0.10^{ab}	77 ± 5^{bc}	1.60 ± 0.15^{ab}	447 ± 88^{b}
Puakō Pond	2	5.29 ± 0.61^{b}	10.62 ± 2.33^{a}	110.32 ± 13.48^a	$\textbf{3.94} \pm \textbf{1.24}^{a}$	$120\pm17^{a\text{-}c}$	1.79 ± 0.50^{ab}	655 ± 90^{a}

Number of observations per location, per parameter:

High Elevation Wells: Salinity = 8, Turbidity = 6, $NO_{3^{-}} + NO_{2^{-}} = 11$, $NH_4 + = 11$, TDN = 6, $PO_4^{3^{-}} = 11$, $H_4SiO_4 = 8$

Mid Elevation Wells: Salinity = 20, Turbidity = 16,
$$NO_{3^-} + NO_{2^-} = 25$$
, $NH_4 + = 25$, $TDN = 16$, $PO_4^{3^-} = 25$, $H_4SIO_4 = 20$

Low Elevation Wells: Salinity = 6, Turbidity = 6, NO₃- + NO₂- = 8, NH₄+ = 8, TDN = 6, PO₄³⁻ = 8, H₄SiO₄ = 6

 $Resort\ Shoreline:\ Salinity = 12,\ Turbidity = 12,\ NO_{3^{-}} + NO_{2^{-}} = 12,\ NH_{4} + = 12,\ TDN = 12,\ PO_{4}^{-3} = 12,\ H_{4}SiO_{4} = 12,\ NO_{4} = 12,\ NO_{$

 $Puak\bar{o} \ Shoreline: \ Salinity = 137, \ Turbidity = 137, \ NO_{3^{-}} + NO_{2^{-}} = 135, \ NH_{4} + = 135, \ TDN = 135, \ PO_{4}^{-3^{-}} = 135, \ H_{4}SiO_{4} = 135, \ H_$

 $Puak\bar{o} \ Ponds: \ Salinity = 6, \ Turbidity = 6, \ NO_{3^{-}} + NO_{2^{-}} = 7, \ NH_{4} + = 7, \ TDN = 6, \ PO_{4}{}^{3_{-}} = 7, \ H_{4}SiO_{4} = 6$

This pattern was observed for $NO_3^-+NO_2^-$ and H_4SiO_4 (Table 2). For NH_4^+ , TDN, and PO_4^{-3-} , concentrations were generally similar among watershed locations, with the exception of resorts' shoreline waters being lower than one of the well water locations (Table 2). Along Puakō's shoreline, the southern and middle regions had the highest nutrient concentrations, with respect to $NO_3^-+NO_2^-$, NH_4^+ , TDN, and PO_4^{-3-} (p < 0.0001). In contrast, H_4SiO_4 concentrations were similar among Puakō's shoreline regions. Nutrient concentrations and salinity were similar between the northern and southern resorts.

3.3. $\delta^{15}N$ in Macroalgae

 δ ¹⁵N macroalgal values were highest in south Puakō compared to the middle and northern regions (Fig. 3; p < 0.0001). Seven stations along Puakō's shoreline had δ ¹⁵N macroalgal values + 7‰ or greater, which is considered to be within the range of values measured for sewage (*reviewed in Wiegner et al.*, 2016). At the northern and southern resorts, δ ¹⁵N macroalgal tissue values were similar to each other, and lower than those observed at many of the Puakō shoreline stations (Fig. 3). Values at the resorts were indicative of NO₃⁻ from upper and lower elevational wells, well fertilized soils and ones under Kiawe trees, and ocean water (Fig. 3).

3.4. SIAR mixing models

Sewage contributions to the NO₃⁻ pool varied among watershed locations (Fig. 4; Table 3). Puakō's anchialine ponds had the largest sewage contribution to the NO₃⁻ pool (36 – 55%), followed by Puakō's shoreline waters (23 – 26%), resort shoreline waters (12 – 17%), low elevational groundwater wells (9 – 20%), and mid-elevational groundwater wells (1 – 4%). Along Puakō's shoreline, ten stations had sewage contributing up to 40% or more (high end of the range) to the shoreline NO₃⁻. All stations were located in south (6 stations) and north (4 stations) Puakō.

3.5. Quantifying water quality impairment caused by OSDS

Dye was observed flowing out of shoreline springs during five of the six dye tracer tests. Groundwater flow rates ranged from 3 to 137 m/d, allowing dye to reach the shoreline between 5 h and 11 d. The type of OSDS (septic tank vs. ATU) did not affect how fast dye reached the shoreline, with ATUs having both the slowest and fastest flow rates. In general, dye appeared at the shoreline sooner in front of homes with drainage fields closer to the shoreline. The only dye tracer test where dye was not observed at the shoreline was from a septic system farthest landward. Here, the drainage field was 122 m from the shoreline; at the other five homes, this averaged 40 m. Additionally, this home was located in the northern part of the neighborhood, where fewer groundwater springs have been observed. We may have missed the spring where the dye emerged, adsorption of the dye to organic matter in the septic tank may have been greater than we anticipated, or we did not sample long enough to capture the dye emerging from the spring (after 16 d, flow rate of < 8 m/d).

Concentrations of FIB and nutrients were similar in front of homes with different OSDS (Fig. 5). The average *Enterococcus* spp. concentration in front of homes with different OSDS all exceeded the HDOH statistical threshold value (Fig. 5 A), with concentrations



Fig. 3. Average $(\pm SE) \delta^{15} N$ (‰) of wild shoreline macroalgae collected at 16 stations (south to north: 1–16) along the Puakō shoreline, and at four adjacent resorts north (17: Mauna Kea, 18: Hapuna Prince) and south (19: Fairmont Orchid, 20: Mauna Lani) of the community on Hawai'i Island. Fertilizer values are from a previous study (Wiegner et al., 2016) on Hawai'i Island. Shaded and outlined background areas represent (average \pm SE) $\delta^{15} NO3$ - of N sources (fertilizer, soil, ocean, high elevation groundwater (GW), low elevation GW, and sewage) measured as part of this and an earlier study at Puakō (Abaya et al., 2018a). Fertilizer values are from a previous study (Wiegner et al., 2016) on Hawai'i Island.



Fig. 4. Average proportional contribution of sewage to the NO_3^- pool in different water types (Puakō's shoreline, resorts' shoreline, Puakō anchialine ponds, low elevation wells, and mid elevation wells) measured in Puakō's watershed, Hawai'i Island. Proportions were estimated using SIAR (v. 4.0). Boxplots illustrate the 95th, 75th, and 50th percentiles from light to dark.

Table 3

Mean \pm SE of δ^{15} N- and δ^{18} O- NO₃⁻ and nutrient concentrations of potential N sources (high elevation groundwater, sewage, and kiawe/fertilized soils) collected in Puakō, Hawai'i Island, as part of this study and a previous one (Abaya et al. 2018a). Sources and nutrients without SE indicate single sample (n=1).

Source	^{δ15} N-NO ₃ ⁻ (‰)	^{δ18} O-NO ₃ ⁻ (‰)	$NO_3^++NO_2^-$ (µmol/L)	NH4 ⁺ (µmol/L)	TDN (µmol/L)	PO4 ³⁻ (µmol/L)	H ₄ SiO ₄ (µmol/L)
Groundwater Sewage Soil	$\begin{array}{c} 4.08 \pm 0.31 \\ 11.73 \pm 1.16 \\ 2.57 \pm 1.74 \end{array}$	$\begin{array}{c} 1.57 \pm 0.44 \\ 14.62 \pm 4.77 \\ 1.56 \pm 2.13 \end{array}$	$\begin{array}{l} 95.27 \pm 2.33 \\ 49.06 \pm 33.90 \\ 4775.26 \pm 3051.69 \end{array}$	$\begin{array}{c} 2.29 \pm 0.89 \\ 4803.99 \pm 1160.31 \\ 446.41 \pm 162.12 \end{array}$	$\begin{array}{c} 102\pm1\\ 2787\pm1602\\ 3\end{array}$	$\begin{array}{c} 2.35 \pm 0.06 \\ 307.21 \pm 47.29 \\ 145.33 \pm 111.10 \end{array}$	$775 \pm 53 \\ 467 \pm 97 \\ 1$

ranging from 0 to 34,330 MPN/100 mL. Similarly, *C. perfringens* concentrations in front of the homes with different OSDS encompassed the standard recommended to HDOH for marine recreational waters of 5 CFU/100 mL (Fujioka et al., 1997), but they ranged from 0 to 27 CFU/100 mL and were not higher than the range reported for non-point source sewage pollution of 10 - 100 CFU/100 mL (Fig. 5B, Fung et al., 2007). δ^{15} N in macroalgae was similar in front of homes with the different OSDS (Fig. 6); they were all within the reported sewage range for Puakō. Nutrient concentrations in front of dye tracer test homes were high. Average TDN concentrations exceeded $100 \,\mu$ mol/L (Fig. 5C), with NO₃⁻+NO₂⁻ comprising most of the TDN. Average TDP concentrations were all greater than 3 μ mol/L (Fig. 5C), with PO₄³⁻ comprising most of the TDP. H₄SiO₄ concentrations did differ in front of these homes, but this difference was most likely from salinity, as H₄SiO₄ is an indicator for fresh groundwater (data not shown). Proportional contributions of sewage to the NO₃⁻ pool in front of homes with different OSDS types varied (Fig. 6). Sewage comprised 25 – 35% of the NO₃⁻ in front of homes with cesspools, 41 – 49% in front of homes with septic tanks, and 12 – 23% in front of homes with ATUS (Fig. 6).

3.6. Assessing proposed sewage treatment upgrades

Conversion of the entire Puakō neighborhood to ATUs would reduce average *Enterococcus* spp. and *C. perfringens* concentrations at the shoreline by 30% and 48%, respectively (Appendix, Table A2). This reduction is dependent on the number of cesspools converted and the efficiency of ATUs to remove FIB. For *Enterococcus* spp., this transition would reduce the percent of stations that exceed the HDOH statistical threshold value from 81% to 69%. For *C. perfringens*, the reduction is greater with ATUs, with only one station exceeding the recommended marine standard of 5 CFU/100 mL (Fujioka et al., 1997). These reductions could be greater if the ATUs used are more effective at removing FIB than estimated in our calculations (50% for *Enterococcus* spp., 68% for *C. perfringens*). A STP would reduce FIB concentrations well below the HDOH standards by specifically targeting the removal of FIB with chlorine, ozone, or ultraviolet treatment.

4. Discussion

4.1. Source of sewage to groundwater

This study successfully used a variety of biological and chemical sewage indicators to determine locations in Puakō's watershed where sewage was entering into groundwater. This information is critical for appropriate management actions to be determined and implemented in targeted locations.

FIB concentrations were generally low in Puako's upper watershed and in the adjacent resorts' shoreline waters, and only



Fig. 5. Box and whisker plots comparing water quality parameters in front of homes with differing onsite sewage disposal systems (OSDS, cesspools n = 4, septic tanks n = 3, Aerobic Treatment Units (ATUs) n = 3) in Puakō, Hawai'i Island. A) *Enterococcus* spp., B) *Clostridium perfringens*, C) Total Dissolved Nitrogen (TDN), and D) Total Dissolved Phosphorus (TDP). Samples were collected three times from June – August 2017. Cesspool data were collected as past of an earlier project, three cesspools were samples seven time, and one was samples 12 times from 2014 to 2016 (Abaya et al., 2018a). Red lines on figure indicate state and recommended fecal indicator bacteria standards for marine recreational waters. Results from one-way ANOVA shown on figure $\alpha = 0.05$.



Fig. 6. Box and whisker plot comparing δ^{15} N macroalgae in front of homes with different onsite disposal systems (OSDS: cesspools, septic tanks, aerobic treatment units (ATU) in Puakō, Hawai'i Island. Results from one-way ANOVA are shown on figure $\alpha = 0.05$.

substantially increased or exceeded HDOH standards in Puako's anchialine ponds, wells, and shoreline waters. This was observed for both Enterococcus spp. and C. perfringens. Enterococcus spp. concentrations in Puako's anchialine ponds, wells, and shoreline waters consistently exceeded the HDOH statistical threshold standard of 130 MPN/100 mL (HDOH, 2014). Epidemiological studies have shown that a swimmer's chance of contracting gastroenteritis is 3.6% at Enterococcus spp. concentrations of 35 MPN/100 mL (reviewed in Fujioka et al., 2015). Our results suggest that swimmers along the Puakō shoreline are at an even greater risk of contracting gastroenteritis. All upper- and mid-elevation wells and resorts' shoreline waters had Enterococcus spp. concentrations below 35 MPN/100 mL. Likewise, C. perfringens concentrations consistently exceeded the recommended standard to HDOH of 5 CFU/ 100 mL for marine recreational waters (Fujioka et al., 1997) in all of the anchialine ponds sampled at Puako, as well as some of the Puako's shoreline stations. These C. perfringens concentrations were all within the range reported for non-point source sewage pollution (Fung et al., 2007). Additionally, MST markers specifically developed to detect Bacteroides bacteria originating from the human gut (HF183 and BacHum) were only detected in Puako's anchialine ponds and shoreline waters. Note, that while we did not have human Bacteroides measurements from the resorts' shoreline waters, our interpretation would not have changed as the Enterococcus spp. and C. perfringens concentrations were very low and indicative of low sewage contamination (Table 1 and Fig. 2). Additionally, a linear relationship between HF183 and BacHum concentrations with risk of contracting gastroenteritis has been established (Boehm et al., 2015); using these equations, the risk of contracting gastroenteritis in Puako's anchialine ponds and shoreline waters ranges from 0% to 43%, with the risk being less than 1% most of the time (Fig. 2C). Note, raw sewage has average values $\log_{10} 3.4$ (SD \pm 0.1) and 5.5 (± 0.1) copies/100 mL for HF183 and BacHum, respectively (Shanks et al., 2010), which are an order of magnitude higher than average values found at Puakō (Table 1). Also, the risk of contracting gastroenteritis could be greater than predicted by the Boehm et al., 2015 model as HF183 and BacHum bacteria markers decay in the environment faster than human-associated viral markers (Ahmed et al., 2019). Together, all of these FIB measurements illustrate that sewage is primarily entering the groundwater at Puako, and not within the upper watershed or at the adjacent resorts, and that there are likely health risks to swimming in these shoreline waters.

While FIB concentrations are used to determine if sewage is present and at levels that could pose a health threat to recreational water users, they do not provide information on the relative contribution of sewage to measured pollution levels. One approach that has successfully overcome this limitation is the combined use of stable isotopes of nitrate (δ^{15} N and δ^{18} O) and mixing models (Wiegner et al., 2016). We utilized this approach to determine the percent contribution of sewage to the NO₃⁻ pool at different locations within Puakō's watershed. We found that sewage contributions to the NO₃⁻ pool varied among watershed locations. Sewage comprised the largest percentage of NO₃⁻ in Puakō's anchialine ponds (36 – 55%) and shoreline waters (23 – 26%) – the two locations with the highest FIB concentrations. Sewage comprised a lower percentage of the NO₃⁻ pool at the resorts' shoreline waters (12 – 17%), low elevation groundwater wells (9 – 20%), and mid-elevation groundwater wells (1 – 4%). These findings support conclusions from the FIB measurements that sewage is entering into the groundwater at Puakō.

In Puakō's effort to transition away from cesspools and septic tanks, determining hotspots of sewage pollution along its shoreline is critical for prioritizing removal locations. The region that consistently had the highest sewage indicator values was south Puakō. Here, nutrient concentrations were highest, δ^{15} N in macroalgal tissue and NO₃⁻ were the most reflective of sewage pollution, and mixing models suggest that up to 52% of the NO₃⁻ is comprised of sewage. Two stations (3 and 4) within south Puakō were previously identified as sewage pollution hotspots (Abaya et al., 2018a). There is also substantial evidence that sewage is present at other locations along the shoreline. Mixing models suggest that sewage comprised more than 40% of the NO₃⁻ at four stations in north Puakō. Also, concentrations of all FIB (*Enterococcus* spp., *C. perfringens*, and human-associated *Bacteroides*) were similar among Puakō's three regions. Additionally, it is likely that δ^{15} N macroalgal tissue values were underestimated due to increased N isotope discrimination (up to 6‰) under high NO₃⁻ concentrations (> 10 µmol/L) (Fujita, 1985; Swart et al., 2014). If the δ^{15} N macroalgal values are adjusted for this discrimination, then algal shoreline values fall within the range reported for sewage (> +7‰, reviewed in Wiegner et al., 2016).

The pervasive presence of sewage along Puakō's shoreline is likely a result of all homes using OSDS in an area with permeable basalt, a high water table, and substantial SGD. Similar H_4SiO_4 concentrations among Puakō's shoreline regions substantiates the ubiquitous presence of SGD in the nearshore waters, which carries sewage from the OSDS to shoreline springs.

4.2. Differences among OSDS types

Dye tracer studies provide irrefutable evidence that sewage from OSDS and STP is entering and contaminating water bodies (HDOH, 1984; Yates, 1985; Glenn et al., 2013). Previous dye tracer studies at Puakō demonstrated that sewage from cesspools reached the shoreline within 9 h to 3 d (4–76 m/d) (Abaya et al., 2018a). Our current study expands these findings by including results from septic tanks and ATUs, with sewage travel times from 5 h to 11 d (3–137 m/d). The type of OSDS (cesspool vs. septic tank vs. ATU) or the presence of a leach field did not affect how fast dye reached the shoreline. In general, dye appeared at the shoreline from homes with drainage fields closer to the shoreline sooner than those further away. The fastest flow rates in this study likely occurred where the leach field was built directly atop a highly-fractured portion of the underlying lava flow. These fractures transport sewage to shoreline springs. Slower flow rates were likely in regions with fewer or smaller fractures in the basalt or a greater thicknesses of soil fill between the drainage field and the shoreline. This information is essential for the Puakō community as it will be used to decide which alternative to cesspools will be pursued to reduce or eliminate sewage from Puakō's nearshore waters.

Water quality was also similar in front of homes with different OSDS types, and values for parameters were suggestive of the presence of sewage. *Enterococcus* spp. concentrations consistently exceeded the HDOH statistical threshold value of 130 MPN/100 mL, with some measurements as high as 34,330 MPN/100 mL. *C. perfringens* concentrations encompassed the standard recommended to HDOH for marine recreational waters of 5 CFU/100 mL (Fujioka et al., 1997), but were not higher than the range reported for

non-point sewage pollution of 10 - 100 CFU/100 mL (Fig. 5B; Fung et al., 2007). δ^{15} N macroalgal values were all within the reported range for sewage at Puakō (Fig. 6; Abaya et al., 2018a). Nutrient concentrations were high, with NO₃⁻ comprising the majority of the TDN concentrations, which exceeded 100 µmol/L. Results from the SIAR mixing model suggest that sewage was a substantial component of the shoreline NO₃⁻ pool in front of homes with different OSDS, ranging from 12% to 49% (Fig. 7). These findings suggest that regardless of the type of OSDS used by homes in Puakō, shoreline waters fronting these homes are contaminated with sewage. Any differences in these parameters that may exist in the sewage effluent of different OSDS is lost by the time the sewage reaches the shoreline because of dispersion and tidal mixing of groundwater before discharge at the shoreline.

4.3. Proposed sewage treatment improvements

Most homes at Puakō use cesspools and septic tanks to dispose of their sewage. Cesspools are underground pits used for temporary storage of sewage, while septic tanks are underground chambers, where solids settle out before the effluent drains into a leach field. Beyond removing solids, neither system treats sewage. Septic systems rely on a thick layer of soil below the leach field for microbial breakdown of sewage before it reaches groundwater, but this is absent at Puakō, where the groundwater is close to the land surface and the substrate is highly permeable fractured basalt instead of soil. Hence, three alternatives to cesspools have been proposed for sewage disposal and treatment in Puakō. They include: 1) converting all cesspools to ATUS, 2) connecting to the existing nearby Mauna Lani STP, or 3) building a STP within Puakō. Each of these solutions treats sewage differently and will have a different outcome for future nearshore water quality at Puakō.

ATU systems collect sewage into an underground container, like septic tanks, where solids settle out. However, ATUs have additional treatment beyond the settling out of the solids, whereas septic tanks do not. The type of additional treatment that occurs within an ATU varies with models and brands, but can include a disinfection component, as well as nutrient removal. The type of ATU proposed for Puakō cycles between aerobic, anaerobic, anoxic, and decanting stages (Aqua Engineering, 2015). This treatment cycle removes organic pollutants and nutrients. Our calculations, based on our water quality measurements, stable isotope mixing models, and published information on the ATU proposed for Puakō, suggests that conversion of the entire neighborhood to ATUs would reduce average *Enterococcus* spp. and *C. perfringens* concentrations at the shoreline by 48%. For *Enterococcus* spp., this transition would reduce the percent of stations that exceed the HDOH statistical threshold value from 81% to 69%. For *C. perfringens*, the reduction is greater, with only one station exceeding the recommended marine standard of 5 CFU/100 mL (Fujioka et al., 1997).

Transporting sewage from homes at Puako to a STP is the other option under consideration. One alternative will transport sewage to the Mauna Lani STP (Fig. 1). At this plant, sewage passes through bar screens and an aerated grit chamber before entering two aerated lagoons (Mauna Lani STP Inc., 2006). At the time of this study, lagoon water was chlorinated and then used for irrigation on a sod farm on the Mauna Lani property (Mauna Lani STP Inc., 2006). The other alternative is for Puakō to build an onsite STP. This plant would contain a screening system, secondary treatment tanks and clarifiers, sand filters, and a UV disinfection system, and would have the capability of producing R-1 quality water, which could be used for irrigation (HDOH, 2002; Aqua Engineering, 2015). Crops irrigated with the R-1 water can reduce the amount of water percolating down to the aquifer, and further reduce any remaining nutrients in the water (Aqua Engineering, 2015). With either STP option, sewage from Puako would be completely removed from Puako's groundwater as it will enter into a sewer system. At Puako, this would mean that water now discharging at shoreline seeps would only be a mixture of groundwater and seawater. However, if the Mauna Lani STP is the alternative selected, sewage that has not undergone pathogen or nutrient removal would still be transported via SGD to the nearshore waters of the Puako-Mauna Lani reef. As of 2017 (the end of our study), the Mauna Lani STP stopped chlorinating lagoon water, and now dispose of it via two on-site injection wells. The injection wells are located just upslope from the southern portion of the Puako community. This is potentially concerning for coral reefs in the area as sewage disposed of via injection wells have resulted in tremendous economic and ecological damage on the Island of Maui (van Beukering and Cesar, 2004; Smith et al., 2005). The use of injection wells as a means of sewage disposal was reviewed by the U.S. Supreme Court (04/23/2020) as a result of the coral reef and water quality conditions in Maui (Maui County v. Hawai'i Wildlife Fund, Sierra Club-Maui Group, Surfrider Foundation, West Maui Preservation Association, 2019). The U.S. Supreme Court ruled that the Clean Water Act requires that sewage injection wells have permits to discharge their effluent if it reaches navigable waters, like the ocean, through groundwater.

5. Conclusions

Use of multiple biological and chemical sewage indicators allowed us to successfully identify where sewage was entering into Puakō's watershed groundwater. Sewage indicator values were greatest within Puakō, and sewage quickly reached shoreline waters, 5 h - 10 d. Sewage travel time to Puakō's shoreline and water quality in front of homes was similar among all OSDS types. These results suggest that the underlying geology on each property is more important in affecting the travel time of the sewage to the shoreline than the OSDS type. Our assessment of water quality impairment caused by OSDS revealed that if all cesspools and septic tanks were converted to ATUs across the entire Puakō neighborhood, FIB concentrations would be reduced, but not to acceptable concentrations. If sewage is treated and disposed of through a STP, sewage would be completely removed from Puakō's groundwater. From a water quality standpoint, both in regards to human and coral reef health, Puakō building its own STP would be best. It will have a higher level of treatment than currently used at the Mauna Lani STP. The plant will also convert the sewage into R-1 quality water, which could be used to irrigate crops that can further reduce any remaining nutrients in the water rather than disposing of it through an injection well (HDOH, 2002; Aqua Engineering, 2015). In conclusion, this study's approach can be adapted to other locations with sewage pollution to identify the location that sewage is entering into the groundwater and to assess potential management actions to improve water



Fig. 7. Proportional contributions of sewage as a NO3- source for three onsite sewage disposal systems (OSDS: cesspool, septic tanks, ATUs)estimated using SIAR (v. 4.0) in Puakō, Hawai'i Island. Boxplots illustrate the 95th, 75th, and 50th percentiles from light to dark.

quality.

CRediT authorship contribution statement

Tracy N. Wiegner: Conceptualization, Data curation, Formal analysis, Funding acquisition, Project administration, Writing – original draft, Writing – review & editing. **Steven L. Colbert:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Project administration, Writing – original draft, Writing – review & editing. **Leilani M.: Abaya:** Data curation, Formal analysis, Project administration, Visualization, Writing – original draft, Writing – review & editing. **Jazmine Panelo:** Data curation, Formal analysis, Writing – review & editing. **Craig E. Nelson:** Formal analysis, Funding acquisition, Project administration, Writing – original draft, Writing – review & editing. **Craig E. Nelson:** Formal analysis, Funding acquisition, Project administration, Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

Thank you to our collaborators: Puakō Community Association, The Nature Conservancy, South Kohala Conservation Action Plan Program Coordinator, Coral Reef Alliance, UH Hilo Analytical Laboratory, C. Edens, E. Stamper, L. Economy, M. Takakusagi S. Adnan Sultan, C. Thompson, C. Garson-Shumay, T. Gerken, Amy Olsen, C. Demapan, J. Stuart, and B. Tonga. This project was supported by funding from: Hawai'i Division of Aquatic Resources Coral Reef Working Group (Grant No. NA15NOS4820037), National Oceanic and Atmospheric Administration (NOAA) Coral Reef Conservation Program (Project No. NA14NOS4820087), UH Hilo Pacific Internship Program for Exploring Science (PIPES, National Science Foundation (NSF) Grant No. 1005186, 1461301), Center for Microbial Oceanography and Education (C-MORE, National Science Foundation Grant No. 0424599NSF), National Science Foundation (NSF) Established Program to Stimulate Competitive Research (EPSCoR,Grant No. OIA-1557349), UH Hilo's Students of Hawai'i Advanced Research Program (SHARP, National Institutes of Health Research Initiative for Scientific Enhancement Award No. R25GM11347), UH Hilo Research Council (Seed Funds, P.I. Wiegner), and the UH Hilo Marine Science Department.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.ejrh.2021.100947.

References

- Abaya, L., Wiegner, T., Beets, J., Colbert, S., Carlson, K., Kramer, L., 2018b. Spatial distribution of sewage pollution on a Hawaiian coral reef. Mar. Pollu. Bull. 130, 335–347.
- Abaya, L., Wiegner, T., Colbert, S., Beets, J., Carlson, K., Kramer, L., Couch, C., 2018a. A multi-indicator approach for identifying shoreline sewage pollution hotspots adjacent to coral reefs. Mar. Pollu. Bull. 129 (1), 70–80.
- Abbott, I.A., 1999. Marine red algae of the Hawaiian Islands. Bishop Museum Press, Honolulu, Hawai'i.
- Abbott, I.A., Huisman, J.M., 2004. Marine Green And Brown Algae of the Hawaiian Islands. Bishop Museum Press, Honolulu, Hawai'i.
- Aguiar, D.K., 2020. Detection and impacts of sewage pollution on South Kohala's coral reefs. M.S. Thesis. Tropical Conservation Biology and Environmental Science Graduate Program. University of Hawai'i at Hilo, Hawai'i.
- Ahmed, W., Zhang, Q., Kozak, S., Beale, D., Gyawali, P., Sadowsky, M.J., Simpson, S., 2019. Comparative decay of sewage-associated marker genes in beach water and sediment in a subtropical region. Water Res. 149, 511–521.
- Aqua Engineering, 2015. Puakō, Hawai'i, Community feasibility study and preliminary engineering report. Prepared for the Coral Reef Alliance.

ASTM D6503-19, 2019. Standard test method for Enterococci in water using Enterolert. ASTM International, West Conshohocken, PA.

- van Beukering, P., Cesar, H.S., 2004. Ecological economic modeling of coral reefs: evaluating tourist overuse at Hanauma Bay and algae blooms at the Kihei Coast, Hawai'i. Pac. Sci. 58 (2), 243–260.
- Bisson, J.W., Cabelli, V.J., 1979. Membrane filter enumeration method for *Clostridium perfringens*. Applie. Environ. Microbiol. 37, 55–66.
- Boehm, A.B., Soller, J.A., Shanks, O.C., 2015. Human-associated fecal quantitative polymerase chain reaction measurements and simulated risk of gastrointestinal illness in recreational waters contaminated with raw sewage. Environ. Sci. Technol. Lett. 2 (10), 270–275.
- Cesar, H., Burke, L., Pet-Soede, L., 2003. The Economics of Worldwide Coral Reef Degradation. Cesar Environmental Economics Consulting.
- Chauret, C., Springthorpe, S., Sattar, S., 1999. Fate of *Cryptosporidium* oocysts, *Giardia* cysts, and microbial indicators during wastewater treatment and anaerobic sludge digestion. Can. J. Microbiol. 45 (3), 257–262.
- Conservation International, 2008. Economic values of coral reefs, mangroves, and seagrasses: a global compilation. Center for Applied Biodiversity Science. Conservation International, Arlington, VA, USA.
- Coplen, T.B., Qi, H., Révész, K., Casciotti, K., Hannon, J.E., 2012. Determination of the δ¹⁵N and δ¹⁸O of nitrate in water, RSIL lab code 2900, chap. 17 of stable isotope-ratio methods, sec. C. In: Révész, K., Coplen, T.B. (Eds.), Geologic Survey Techniques and Methods, 10. Methods of the Reston Stable Isotope Laboratory, U.S., p. 35
- Couch, C.S., Most, R., Wiggins, C., Minton, D., Conklin, E., Sziklay, J., Amimoto, R., Pollock, K., Caldwell, Z., 2014. Understanding the consequences of land-based pollutants on coral health in South Kohala. The Nature Conservancy. 41 p.
- Derse, E., Knee, K.L., Wankel, S.D., Kendall, C., Berg Jr, C.J., Paytan, A., 2007. Identifying sources of nitrogen to Hanalei Bay, Kaua'i, utilizing the nitrogen isotope signature in macroalgae. Environ. Sci. Technol. 41, 5217–5223.
- Dudley, B., Hughes, F., Ostertag, B., 2014. Groundwater availability mediates the ecosystem effects of an invasion of Prosopis pallid. Ecol. Appl. 24, 1954–1971.
- Elmir, S., (2018). Septic systems vulnerable to sea level rise. Final report to Board of County Commissioners, Miami-Dade County, in support of resolution no. R-911-16.
- Engott, J.A., 2011. A water-budget model and assessment of groundwater recharge for the Island of Hawaii. USGS Scientific Investigations Report 2011-5078.
- Fujioka, R., Hashimoto, H., Siwak, E., Young, R., 1981. Effect of sunlight on survival of indicator bacteria in seawater. Applie. Environ. Microbiol. 41, 690–696.
 Fujioka, R.S., Roll, B., Byappanahalli, M., 1997. Appropriate recreational water quality standards for Hawai'i and other tropical regions based on concentrations of *Clostridium perfringens*. Proc. Water Environ. Fed. 4, 405–411.
- Fujioka, R., Solo-Gabriele, H., Byappanahalli, M., Kirs, M., 2015. US recreational water quality criteria: a vision for the future. Int. J. Env. Res. Public Health 12 (7), 7752–7776.
- Fujita, R.M., 1985. The role of nitrogen status in regulating transient ammonium uptake and nitrogen storage by macroalgae. J. Exp. Mar, Biol. Ecol. 92, 283–301. Fukunaga Assoc. Inc., 2010. Hawai'i County Water Use and Development Plan Update. Final Report. 803, 1–34.
- Fung, D.Y.C., Fujioka, R., Vijayavel, K., Sato, D., Bishop, D., 2007. Evaluation of Fung Double Tube Test for Clostridium perfringens and Easyphage test for F-specific RNA coliphages as rapid screening tests for fecal contamination in recreational waters of Hawai'i. J. Rapid Meth. Aut. Mic. 15, 217–229.
- Giambelluca, T.W., Chen, Q., Frazier, A.G., Price, J.P., Chen, Y.-L., Chu, P.-S., Eischeid, J.K., Delparte, D.M., 2013. Online rainfall atlas of Hawai'i. Bull. Amer. Meteor. Soc. 94 (3), 313–316. https://doi.org/10.1175/BAMS-D-11-00228.1.
- Glenn, C.R., Whittier, R.B., Dailer, M.L., Dulaiova, H., El-Kadi, A.I., Fackrell, J., Kelly, J.L., Waters, C.A., Sevadjian, J., 2013. Lahaina groundwater tracer study, Lahina, Maui, Hawai'i. Report prepared for State of Hawai'i Department of Health, U.S. Environmental Protection Agency, U.S. Army Engineer Research and Development Center.
- HDAR, South Kohala reefs in dire straits Public informational sheet, 2013.
- HDOH, Interim report of a baseline study on water quality at Kapoho Bay, Hawai'i, 1984.
- HDOH, Guidelines for the treatment and use of recycled water Report prepared by Hawai'i State Department of Health Wastewater Branch 2002.
- HDOH, State of Hawai'i water quality monitoring and assessment report HDOH CWA §305(b), 2014.
- HDOH, 2015. Rationale for the proposed revisions to Department of Health Administrative Rules, Title 11. Chapter 62 Wastewater Systems. Department of Health Environmental Management Division Wastewater Branch Honolulu, Hawai'i, p. 2015 (July).
- HDOH, 2017. Report to the 29th legislature State of Hawai'i 2018 regular session relating to cesspools and prioritization for replacement. Report in response to Act 125, 2017 Regular Session (House Bill 1244, HD1, SD2, CD1).
- Hunter, C., Evans, C., 1995. Coral reefs in Kāne'ohe Bay, Hawai'i: Two centuries of western influence and two decades of data. Bull Mar Sci 57, 501–515. Knowlton, N., Brainard, R.E., Fisher, R., Moews, M., Plaisance, L., Caley, M.J., 2010. Coral reef biodiversity. In: McIntyre, A.D. (Ed.), Life in the World's Oceans:
- Diversity Distribution and Abundance. Blackwell Publishing Ltd, Oxford, U.K, pp. 65–74.
- Lapointe, B., Barile, P., Wynne, M., Yentsch, C., 2005. Reciprocal *Caulerpa* invasion: mediterranean native *Caulerpa* ollivieri in the Bahamas supported by human nitrogen enrichment. Aquat. Invaders 16, 1–5.
- Mauna Kea Properties Inc. Before the land use commission of the State of Hawai'i Docket A84-574 1985 1 27.
- Mauna Lani STP Inc. Before the public utilities commission of the State of Hawai'i. Docket No. 05-0229 Decision and Order No. 22299 2006 1 41.
- McKenzie, T., Habel, S., Dulai, H., 2021. Sea-level rise drives wastewater leakage to coastal waters and storm drains. Limnol. Oceangr. Lett. 121, 800. https://doi.org/ 10.1002/lol2.10186.
- Mink , J.F., Lau, L.S. , WRRCTR No. 191 Aquifer identification and classification for the island of Hawai'i Groundwater protection strategy for Hawai'i, 1993.
- Minton m D., Conklin, E., Weiant, P., Wiggins, C., Forty years of decline on Puakō's coral reefs A review of historical and current data (1970-2010) The Nature Conservancy, Honolulu, HI, 2012 1 140.
- NOAA NOAA Habitat Blueprint West Hawai'i Implementation Plan, 2016 1 54.
- Parnell, A.C., Inger, R., Bearhop, S., Jackson, A.L., 2010. Source partitioning using stable isotopes: Coping with too much variation. PLoS One 5 (3), e9672. https:// doi.org/10.1371/journal.pone.0009672.
- Parsons, M.L., Walsh, W.J., Settlemier, C.J., White, D.J., Ballauer, J.M., Ayotte, P.M., Osada, K.M., Carman, B., 2008. A multivariate assessment of the coral ecosystem health of two embayments on the lee of the island of Hawai'i. Mar. Pollut. Bull. 56, 1138–1149.
- Pastorok, R., Bilyard, G., 1985. Effects of sewage pollution on coral-reef communities. Mar. Ecol. Prog. Ser. 21, 175–189.
- Paytan, A., Shellenbarger, G.G., Street, J.H., Gonneea, M.E., Davis, K., Young, M.B., Moore, W.S., 2006. Submarine groundwater discharge: An important source of new inorganic nitrogen to coral reef ecosystems. Limnol. Oceanogr. 51, 343–348.
- Peterson, R.N., Burnett, W.C., Glenn, C.R., Johnson, A.G., 2009. Quantification of point-source groundwater discharges to the ocean from the shoreline of the Big Island, Hawai'i. Limnol. Oceanogr. 54, 890–904.

- Pinto, B., 1999. Characterization of faecal streptococci as indicators of faecal pollution and distribution in the environment. Lett. Appl. Microbiol. 29, 258–263.
 Prouty, N.G., Cohen, A., Yates, K.K., Storlazzie D., C., Swarzenski, P.W., White, D., 2017. Vulnerability of coral reefs to bioerosion from land-based sources of pollution. J. Gr. Res. Oceans 122, 9319–9331.
- Redding, J.E., Myers-Miller, R.L., Baker, D.M., Fogel, M., Raymundo, L.J., Kim, K., 2013. Link between sewage-derived nitrogen pollution and coral disease severity in Guam. Mar. Pollut. Bull. 73 (1), 57–63.
- Schott Mauna Lani, E., DW]A⁻ina Le]a and Puako⁻/Wailea Bay wastewater assessment Professional Report to the Puako⁻ Community Association Phase One 2010 1 47.
- Shanks, O.C., White, K., Kelty, C.A., Sivaganesan, M., Blannon, J., Meckes, M., Varma, M., Haugland, R.A., 2010. Performance of PCR-based assays targeting Bacteroidales genetic markers of human fecal pollution in sewage and fecal samples. Environ. Sci. Technol. 44 (16), 6281–6288.
- Sharp, J.H., Rinker, K.R., Savidge, K.B., Abell, J., Benaim, J.Y., Bronk, D., Burdige, D.J., Cauwet, G., Chen, W., Doval, M.D., Hansell, D., 2002. A preliminary methods comparison for measurement of dissolved organic nitrogen in seawater. Mar. Chem. 78 (4), 171–184.
- Shuval, H., 2003. Estimating the global burden of thalassogenic diseases: Human infectious diseases caused by wastewater pollution of the marine environment. J. Water Health 1, 53–64.
- Smith, S.V., Kimmerer, W.J., Laws, E.A., Brock, R.E., Walsh, T.E., 1981. Kāne'ohe Bay sewage diversion experiment: Perspectives on ecosystem responses to nutritional perturbation. Pac. Sci. 35 (4), 279–395.
- Smith, J.E., Runcie, J.W., Smith, C.M., 2005. Characterization of a large-scale ephemeral bloom of the green alga *Cladophora sericea* on the coral reefs of West Maui, Hawai'i. Mar. Ecol. Progr. Ser. 302, 77–91.
- Sutherland, K.P., Porter, J.W., Turner, J.W., Thomas, B.J., Looney, E.E., Luna, T.P., Meyers, M.K., Futch, J.C., Lipp, E.K., 2010. Human sewage identified as likely source of white pox disease of the threatened Caribbean elkhorn coral, *Acropora palmata*. Environ. Microbiol. 12 (5), 1122–1131.
- Swart, P.K., Evans, S., Capo, T., Altabet, M.A., 2014. The fractionation of nitrogen and oxygen isotopes in macroalgae during the assimilation of nitrate.

Biogeosciences 11, 6147–6157.

UNESCO, Background papers and supporting data on the practical salinity scale 1978 UNESCO technical papers in marine science, 1981 37.

USEPA, 2011. Part 136, Appendix B- Definition and procedure for the determination of the method detection limit- revision 1.11. Code of Federal Regulations. Government Printing Office.

Wear, S., Vega Thurber, R., 2015. Sewage pollution: mitigation is key for coral reef stewardship. Ann. N. Y. Acad. Sci. 1355, 1355-30.

- Whittier, R., El-Kadi, A., Human health and environmental risk ranking of on-site sewage disposal systems for the Hawaiian islands of Kaua'i, Moloka'i, Maui, and Hawai'i 2014. http://health.hawaii.gov/wastewater/files/2015/09/OSDS_NLpdf.
- Wiegner, T., Mokiao-Lee, A., Johnson, E., 2016. Identifying nitrogen sources to thermal tide pools in Kapoho, Hawai'i, U.S.A, using a multi-stable isotope approach. Mar. Pollu. Bull. 103 (1–2), 63–71.
- Withers, P.J.A., Jarvie, H.P., Stoate, C., 2011. Quantifying the impact of septic tank systems on eutrophication risk in rural headwaters. Environ. Int. 37 (3), 644–653. Wolfe, E.W., Morris, J., 1996. Geologic map of the Island of Hawai'i: U.S. Geological Survey Miscellaneous Investigations Series I–2524-A, scale 1:100,000. Yates, M.V., 1985. Septic tank density and groundwater contamination. Groundwater 23 (5), 586–591.
- Yoshioka, R.M., Kim, C.J.S., Tracy, A.M., Most, R., Harvell, C.D., 2016. Linking sewage pollution and water quality to spatial patterns of *Porites lobata* growth anomalies in Puakō, Hawai'i. Mar. Pollut. Bull. 104, 313–321.