URTeC: 2900727

Environmental Impact Analysis on the Hydraulic Fracture Test Site (HFTS)

Sarah Eisenlord*[1], Thomas Hayes[1], Kent Perry[1]; 1. Gas Technology Institute, Des Plaines, IL

This paper was prepared for presentation at the Unconventional Resources Technology Conference held in Houston, Texas, USA, 23-25 July 2018.

Abstract

Objective

The Hydraulic Fracture Test Site (HFTS) provided a platform to test new extraction and production techniques and technologies to understand fracture geometry and increase the efficiency of shale hydrocarbon production. Eleven test wells were drilled in the Upper and Middle Wolfcamp formations, hydraulically fractured, and produced. Air quality, groundwater quality, and produced water chemistry and microbiology were monitored through the duration of the field experiment and one year into production.

Methods

BTEX, methane, VOCs, SOx, NOx, and PM_{10} were measured 1,000 ft up-wind and 1,000 ft down-wind of the HFTS experiment before operations began, during hydraulic fracturing, flowback, and several months into production to evaluate operational impact on local air quality. The underlying Edward-Trinity Plateau aquifer was monitored using 5 groundwater wells within 2 miles of the HFTS pad; BTEX, methane, petroleum hydrocarbons, and a comprehensive suite of anions and metals were analyzed from samples taken before operations began, during hydraulic fracturing, flowback, and one year after production. Water produced from 3 of the 11 HFTS wells was analyzed for chemical and microbiological properties 1.5 years into production.

Observations

There was no significant impact to air quality from HFTS operations, although elevated BTEX concentrations were measured 1,000 ft. away from the well-pads during the flowback period. We detected no migration of hydrocarbon or fracture fluid chemical migration into the Edward-Trinity Plateau aquifer. Source waters used for hydraulic fracturing contained naturally occurring high concentrations (~500ppm) of sulfate. Rapid microbially induced corrosion was detected in several wells, leading to biocide dosing. Microbiological populations were significantly altered by biocide treatment, but still continued to increase in population size six months after application.

Significance

After hydraulic fracturing, detailed chemical and microbiological analysis provides evidence for microbiologically-mediated sub-surface transformations of supplied sulfate and organic carbon into sulfides. This report discusses air quality, water quality, and produced water analytical results for the HFTS experiment and further implications for the unconventional oil and gas industry.

Introduction

Hydrocarbons derived from shale deposits in the United States are poised to become instrumental in our energy independence and a clean energy future. These resources are projected to constitute over 38% of the national hydrocarbon energy portfolio by 2040. The Permian Basin is the largest petroleum-producing basin in the United States and has produced a cumulative 28.9 billion barrels of oil and 75 trillion cu ft. of gas. Currently, nearly 2 million barrels of oil a day are being produced from the basin. Constant innovations in horizontal drilling and hydraulic fracturing technologies are increasing shale hydrocarbon production efficiency and minimizing environmental impact.

Sustainable production of these resources requires a clear understanding of the potential environmental impacts arising from shale development as well as effective strategies for managing risks to the public health and the environment (Small, et al., 2014). A number of reviews in the literature have identified various areas of risk and have discussed pathways of potential impacts to the environment (Small, et al., 2014; USEPA, 2016; Clark, et al., 2013). With regard to public opinion and regulatory interaction, significant concerns have been raised over the environmental impacts of shale development on groundwater and ambient air receptors. These concerns arise from an awareness of the large amounts of water that are handled and of the substantial mechanized equipment (e.g. diesel driven pumps) and high pressures that are employed in shale well field development.

Economic recovery of shale resources is dependent on the pressurized injection of large volumes of chemically treated water to hydraulically fracture the shale and alter its permeability through the generation of extensive fracture networks. As horizontal wells become longer and fracture treatments more complex, water use and management needs will increase. The practice of produced water recycling and re-use substantially decreases the industry's reliance on fresh water sources and disposal needs, yet may introduce new and unforeseen risks to infrastructure integrity and formation productivity through the re-introduction of sub-surface tolerant microorganisms. Biogeochemical processes carried out by microorganisms in the deep subsurface has been of much interest to microbiologists and geochemists since shale hydrocarbon extraction began to expand in the United States (Magot, 2000; Roling, 2003; Voordouw, 2009; Mohan, 2013; Strutchtemeryer, 2012; Daly, 2016; Lipus, 2017).

There is widespread evidence of microbial growth with complex metabolic abilities in shale petroleum reservoirs across the United States (Magot, 2000; Galperin, 2011; Gaspar J., 2014; Daly, 2016). Potential microbial mechanisms include (1) increased methane production from microbially produced hydrogen and carbon dioxide or acetate, (2) biodegradation of crude oil, (3) biomass accumulation leading to bio-plugging of fractures or surface equipment, (4) iron sulfide deposition decreasing permeability, (5) corrosion of steel well infrastructure, and (6) production of sour gas. Despite the potential impacts, little is known about the function, activity, and controls of microorganisms in hydraulically fractured shale. Key questions remain regarding the introduction and proliferation of microorganisms in the subsurface, mechanisms for mitigating deleterious impact, and how production operations influence the subsurface environment regarding microbial activity.

The aim of this study was to conduct careful measurements of constituents of concern in ambient air and groundwater strata proximal to shale gas well construction, completion, and production associated with the HFTS in the Permian Basin, TX. The environmental monitoring program focused on determining the condition of the groundwater and ambient air before, during, and after critical shale well actions that were performed at the HFTS.

Additionally, the research presented in this report is aimed to illuminate microbial population dynamics in fracture fluids, produced water (over one year of production) and treated, recycled produced water all at the HFTS. The HFTS experiment was designed to test new extraction and production techniques and technologies aimed at increasing the efficiency of shale hydrocarbon production. Eleven test wells were drilled in the Wolfcamp Formation, hydraulically fractured, and produced. This study examines the microbiological populations in three of the eleven wells.

Description of the Hydraulic Fracture Test Site

Situated about 55 miles southeast of Midland, TX, the HFTS, comprised of oil and gas assets owned and operated by Laredo Petroleum, is located in the Northeast quadrant of Reagan County near the northern boundary of the county (**Figure 1**). The test site included eleven wells; six horizontal wells drilled into the Upper Wolfcamp play and five horizontal wells drilled into the Middle Wolfcamp in a chevron pattern. Five groundwater wells, within a 2 mile radius of the drill pads, supply the site with water necessary for hydraulic fracturing from the Edward-Trinity Plateau aquifer. Laredo owns and operates an on-site facility for produced water recycling and re-use.

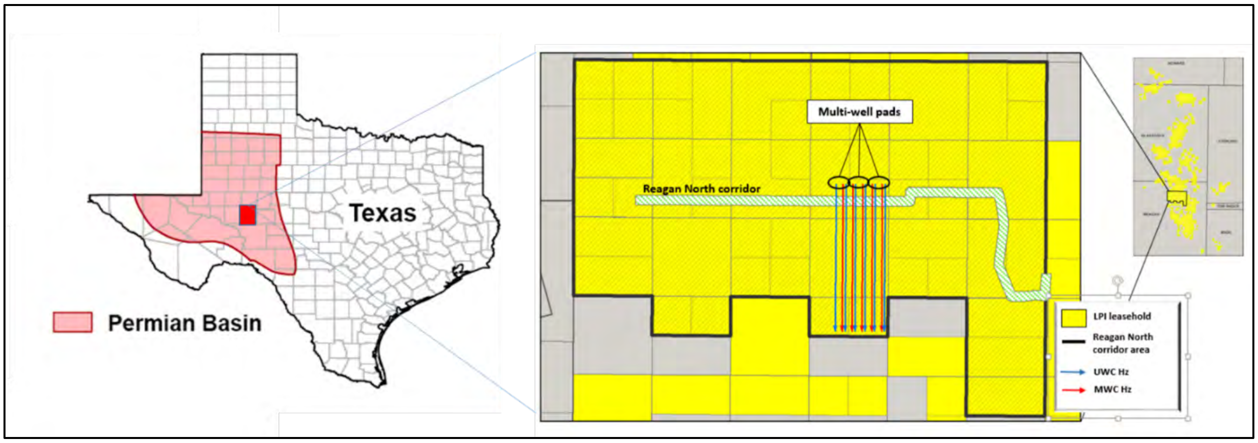


Figure 1: Location of the Hydraulic Fracture Test Site in Reagan, TX, USA

Methods

Groundwater Monitoring

Five groundwater wells were sampled throughout the HFTS experiment to measure the impact of development activities on groundwater reserves. Water was collected at five time points: (1) baseline (before hydraulic fracturing), (2) during hydraulic fracturing, (3) during flowback, (4) six months into production, and (4) one year into production. The wells ranged 350 to 740 feet deep. Water was collected directly from the well head using the direct fill method. The list of parameters measured at each sampling point and the methods used are given in **Table 1.** TOC and BTEX were analyzed at Test America in Northwest Chicago, IL, all other analyses were conducted at the Gas Technology Institute in Des Plaines, IL (GTI).

Table 1: Parameters and Methods for Groundwater Quality Analysis at the HFTS

Parameter	Sample Preservation	Standard Method	Instrumentation	Comments
General Chemistry				
pH	Ice	EPA 9040	Electrometric Equipment	Suitable for water analysis
Total Dissolved Solids	Ice	SM18 2540 C	Gravimetric Apparatus	Standard USEPA Method
Specific Conductance	Ice	EPA 120.1	Self-Contained Conductivity Meter	Standard USEPA Method
Alkalinity	Ice	SM18 2320 B	Titration Equipment	21st Ed. Standard Methods (2015)
Organic Compounds				
Total Organic Carbon	Sulfuric Acid	EPA 415.1	Combustion and Infrared Detection	Provides the concentration of total organics as Carbon.
Dissolved Methane	Sulfuric Acid	EPA RSK SOP 175	Gas Chromatography with thermal conductivity, flame ionization or electron capture detector	Uses purge and trap sample preparation procedures.
Volatile Organic Compounds	Hydrochloric Acid	EPA 8260B or 8021B	Gas Chromatography with Mass Spectroscopy or	Benzene, Toluene, Ethylbenzene, Xylenes

			Photoionization and Flame Ionization Detection	
Inorganic Species				
Cations	Nitric Acid	EPA 200.7	Inductively Coupled Plasma Emission Spectroscopy	As, Ba, B, Cd, Ca, Cr, Cu, Fe, Pb, Mg, Mn, Ni, K, Na, Zn
Anions	Ice	EPA 300.0	Ion Chromatography	Chloride, Fluoride, Sulfate, Nitrate
		EPA 9030B (modified)	Distillation / Ion Chromatography	Total Sulfide

Air Quality Analysis

The air monitoring effort was designed to determine the effect of shale development operations on the quality of ambient air at ground level and at distances proximal to shale energy well pads.

A Vantage Pro2 (Davis Instruments) measured the prevailing wind direction and speed during the experiment to determine placement of air sampling equipment 1,000ft upwind and 1,000ft downwind of the center HFTS pad. Samples for BTEX and Methane were collected with a 6L Summa Can with 24-hour valving, volatile organic carbon (VOC), hydrogen sulfide, and PM10 were detected with direct read data loggers (RaePPB, ToxiRae Pro and TSI Dustrak DRX 8533 respectively). Samples for nitrogen dioxide and nitric oxide measurement were taken with a pump and treated sorbent tube for visible absorption spectrophotometry. Sampling occurred at four time-points for 72 consecutive hours. Time points are (1) baseline (before hydraulic fracturing), (2) during hydraulic fracturing, (3) during flowback, and (4) three months into production. The sampling was repeated in Midland, TX at one 72 hour time-point in May for comparative analysis.

Produced Water Chemistry and Microbiology

The overall purpose of the effort to characterize waters associated with the hydraulic fracturing and production at the HFTS was to obtain information of value for long-term produced water management (including reuse) and to gain some insight into possible transformations that may have implications for production, corrosion, and sour gas generation.

The scope of this effort included the collection of samples from:

- Three open top water impoundments that stored freshwater and recycled produced water for hydraulic fracturing operations; sampled weekly for five consecutive weeks.
- Fracture fluids (an 80/20 mix of fresh and recycled produced water) mixed with fracture treatment chemicals; sampled on the first, sixth, and final day of hydraulic fracturing of three wells.
- A time series of produced water samples to characterize the produced water from three selected shale wells. Sampling occurred at days 0, 1, 5, 14, 36, 90, 210, 365, and 550 days following the initial release of flowback water.

More than 10 million gallons of fracture fluids was injected in the course of stimulating each of the shale wells. Post stimulation, the wells were shut in and fracture fluids were held downhole for about six weeks before the initiation of flowback release and production.

Water chemistry sampling and analyses for produced water were identical to the groundwater sampling with the addition of microbiological analyses **(Table 1)**. Water for microbiological analyses was collected in two 1L sterile Nalgene bottles and kept cold at 4°C until overnight shipment on ice to GTI within 4 days of collection. Water samples were vacuum filtered through 0.2um PES filters upon arrival and filters were immediately frozen at -80°C until DNA extraction.

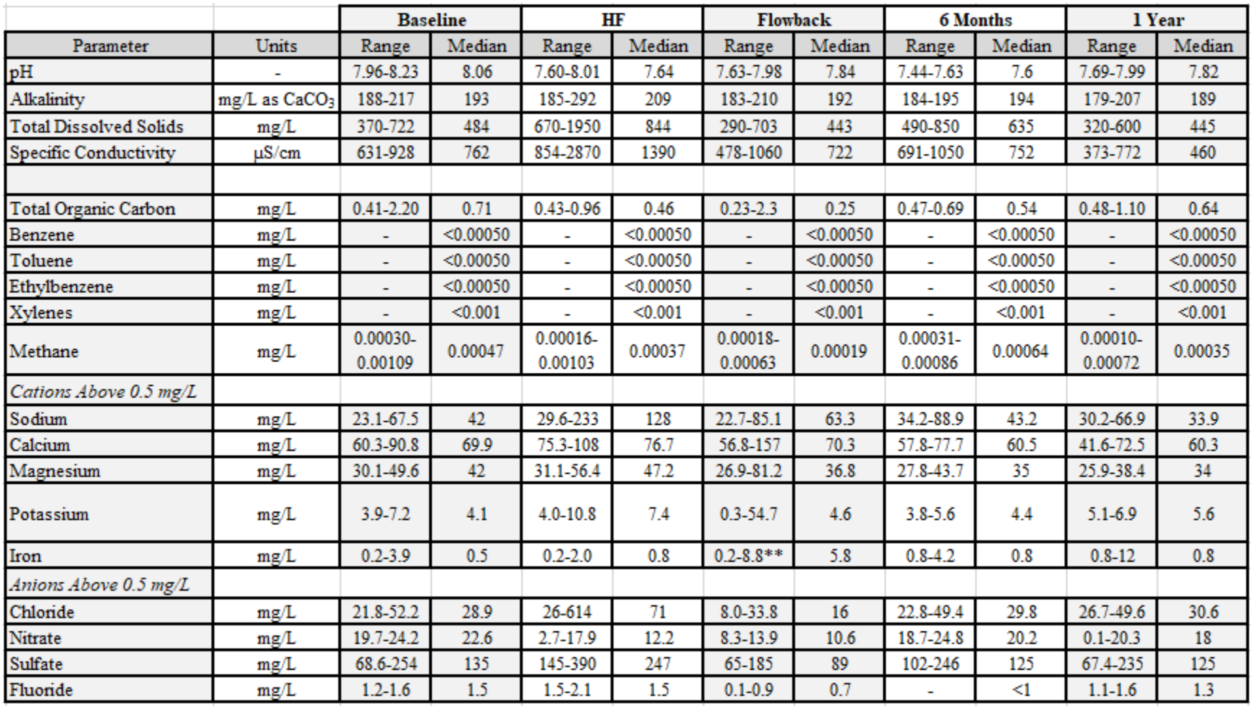
Microbial DNA was extracted from the frozen filters with an optimized protocol using the MP Biomedicals FastDNA Spin Kit for Soil DNA Extraction Kit. Extracted DNA was quantified with the Qubit (ThermoFisher), and aliquoted for archive, qPCR analysis, and 16S rRNA gene sequencing. Quantitative PCR (qPCR) was conducted at GTI on 1:10 dilutions of extracted DNA according to the NACE TM0106-2006 and TM0212-2012 standards. Genes targeted with qPCR include 16S RNA to estimate total Bacteria, and functional genes for sulfate reduction, methane production, and nitrate reduction.

Results and Discussion

Groundwater Quality

Overall, the data from the groundwater monitoring indicates there was little impact on groundwater quality measured as a consequence of HFTS operations. The data presented in **Table 2** show that during the baseline and throughout the sampling campaign, the quality of the groundwater remained high quality, suitable for residential use. Generally, the water was low in total dissolved solids at median values less than 800 mg/L. Calcium concentrations ranged from 22.7 to 157 mg/L, concentrations greater than 60 mg/L categorized the groundwater as “hard”. The ratio of sodium and calcium/ magnesium cations was sufficient to maintain the sodium absorption ratio (SAR*) at values well below 6, which makes the water highly suitable for irrigation of crops. Only five of the fifteen metals sought after in the analyses of the water samples were consistently present in concentrations over 0.5 ppm. The five cations (sodium, calcium, magnesium, potassium, and iron) that were greater than 0.5 ppm are included in **Table 2.**

Table 2: Groundwater Quality Results during the HFTS Experiment



		Baseline		HF		Flowback		6 Months		1 Year	
Parameter	Units	Range	Median	Range	Median	Range	Median	Range	Median	Range	Median
pH	-	7.96-8.23	8.06	7.60-8.01	7.64	7.63-7.98	7.84	7.44-7.63	7.6	7.69-7.99	7.82
Alkalinity	mg/L as $CaCO_3$	188-217	193	185-292	209	183-210	192	184-195	194	179-207	189
Total Dissolved Solids	mg/L	370-722	484	670-1950	844	290-703	443	490-850	635	320-600	445
Specific Conductivity	µS/cm	631-928	762	854-2870	1390	478-1060	722	691-1050	752	373-772	460
Total Organic Carbon	mg/L	0.41-2.20	0.71	0.43-0.96	0.46	0.23-2.3	0.25	0.47-0.69	0.54	0.48-1.10	0.64
Benzene	mg/L	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050
Toluene	mg/L	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050
Ethylbenzene	mg/L	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050	-	<0.00050
Xylenes	mg/L	-	<0.001	-	<0.001	-	<0.001	-	<0.001	-	<0.001
Methane	mg/L	0.00030-0.00109	0.00047	0.00016-0.00103	0.00037	0.00018-0.00063	0.00019	0.00031-0.00086	0.00064	0.00010-0.00072	0.00035
Cations Above 0.5 mg/L											
Sodium	mg/L	23.1-67.5	42	29.6-233	128	22.7-85.1	63.3	34.2-88.9	43.2	30.2-66.9	33.9
Calcium	mg/L	60.3-90.8	69.9	75.3-108	76.7	56.8-157	70.3	57.8-77.7	60.5	41.6-72.5	60.3
Magnesium	mg/L	30.1-49.6	42	31.1-56.4	47.2	26.9-81.2	36.8	27.8-43.7	35	25.9-38.4	34
Potassium	mg/L	3.9-7.2	4.1	4.0-10.8	7.4	0.3-54.7	4.6	3.8-5.6	4.4	5.1-6.9	5.6
Iron	mg/L	0.2-3.9	0.5	0.2-2.0	0.8	0.2-8.8**	5.8	0.8-4.2	0.8	0.8-12	0.8
Anions Above 0.5 mg/L											
Chloride	mg/L	21.8-52.2	28.9	26-614	71	8.0-33.8	16	22.8-49.4	29.8	26.7-49.6	30.6
Nitrate	mg/L	19.7-24.2	22.6	2.7-17.9	12.2	8.3-13.9	10.6	18.7-24.8	20.2	0.1-20.3	18
Sulfate	mg/L	68.6-254	135	145-390	247	65-185	89	102-246	125	67.4-235	125
Fluoride	mg/L	1.2-1.6	1.5	1.5-2.1	1.5	0.1-0.9	0.7	-	<1	1.1-1.6	1.3

Organic compounds were low in all water samples taken at all sampling times from the five-groundwater wells. Total organic carbon remained less than 1 mg/L and the sentinel compounds for impact from shale wells including benzene, toluene, and ethylbenzene that remained less than 0.5 ppb, and xylenes that remained less than 1 ppb. Median values for methane, the most mobile compound of interest in subsurface strata, remained below 1 ppb providing evidence that no mass transfer of constituents from the shale strata or the producing wells to the overlying groundwater aquifer was occurring.

Significant, measureable changes were observed with two parameters – total dissolved solids (TDS; p-value < 0.01) and conductance (p-value < 0.01) – during one time period, hydraulic fracturing (**Figure 2**). The median value for TDS increased by about 75% from 484 to 844 mg/L from the baseline to the hydraulic fracturing time period; subsequently, median TDS values recovered to baseline levels in the 1 to 10 months that followed. Even at the elevated salinity levels, median TDS levels remained less than 850 mg/L, far below the level of TDS (3,000 mg/L) that defines the highest record TDS from the Edward-Trinity Plateau aquifer (Anaya & Jones, Texas Water

Development Board, 2009). The median value for conductance among the five-groundwater wells showed an increase from 762 to 1,390 µS/cm from the Baseline to the Hydraulic Fracturing stage, and subsequently recovered to Baseline values.

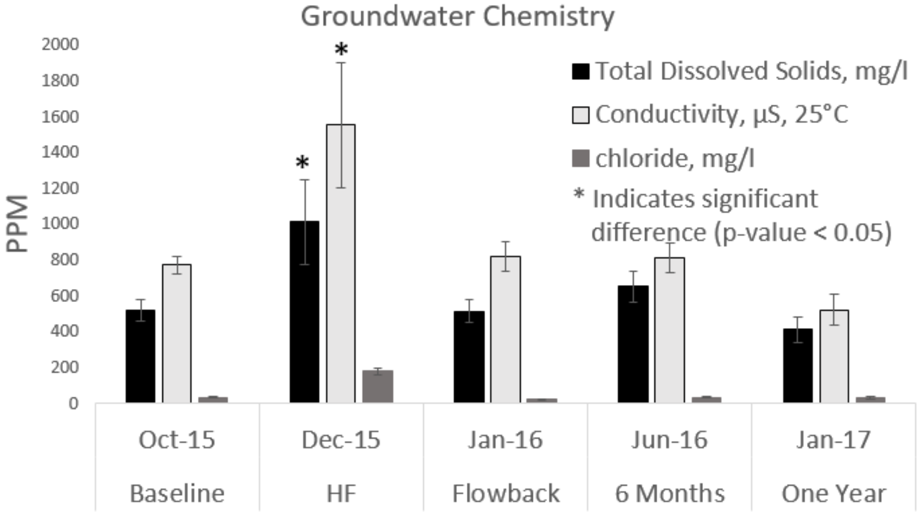


Figure 2: Groundwater Salinity Changes during HFTS Experiment

The impact on salinity of the groundwater was detectable only during the stage of operations when hydraulic fracturing of the 11 wells was being performed and large volumes of water were being withdrawn from the local aquifer to provide water for hydraulic fracturing. Since the rise of salinity was not accompanied with any trace of methane or BTEX, it is unlikely that the measured rise in salt in the groundwater was due to water infiltration from the shale strata or from the shale well structures. Preparation for hydraulic fracturing operations involved the pumping of tens of millions of gallons from the groundwater wells into the open top impoundments. Extensive pumping of the wells likely caused considerable drawdown of the aquifer which could have promoted the infiltration of water from lower portions of the aquifer that may have had a higher salinity level. This mechanism is further supported by the rapid return of TDS and conductance levels to baseline values in water samples upon cessation of large withdrawals. Salinity increases in wells due to drawdown is a phenomenon that has been described elsewhere (USEPA, 2016). Additionally, downhole deformation measurements conducted by Haliburton with three arrays of tiltmeters at the test site found no microcosmic events were detected shallower than 6,000ft true vertical depth, several thousand feet below the deepest reaches of the Edward-Trinity Plateau aquifer.

Air Quality

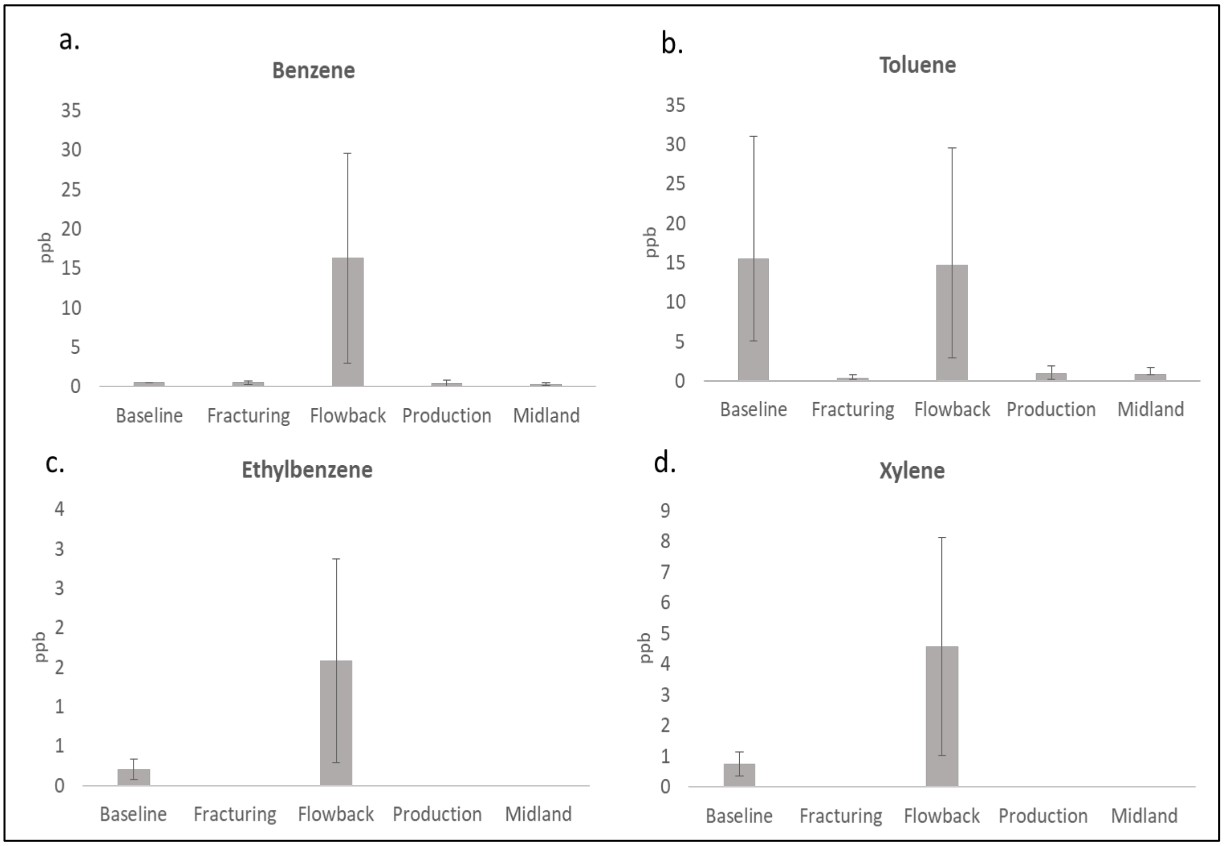
Air monitoring results of the HFTS field experiment during the four phases of activity, including baseline and Midland TX sampling events, are summarized in **Table 3**. Table 3 presents average concentrations (12 or 24 hour) for each parameter, calculated from both upwind and downwind measurements. Upwind and downwind measurements were combined due to the lack of a constant wind direction for each 72-hour sampling effort and the limitation of having only two monitoring stations per sampling. Therefore, this data represents the concentrations of key air quality parameters 1,000 feet away from the oil and gas development site, HFTS. Not included in this table were constituents that were consistently not detected: nitrogen dioxide, nitric oxide and hydrogen sulfide. It should be noted, no nitrogen oxide or nitrogen dioxide was detected at the sampling locations at any of the sampling time points, most notably during hydraulic fracturing when multiple diesel engines were running to pressurize fluids for hydraulic fracturing. This indicates the methods deployed may have been inadequate to accurately measure all emissions at the HFTS field site. Regardless, the data collected are presented and discussed below.

Table 3: Air Quality Results for HFTS Experiment

Background	Units	Average	Max	Min	Std error
Benzene	ppb	0.478	0.570	0.380	0.034
Ethylbenzene	ppb	0.206	0.550	0.000	0.127
Toluene	ppb	15.53	56.00	0.390	10.51
Xylenes	ppb	0.752	1.800	0.000	0.394
Methane	ppm	2.720	3.000	2.500	0.192
PM 10	mg/m3	0.786	2.320	0.014	0.594
VOC	ppb	68.33	205.0	0.000	52.93
Fracturing	Units	Average	Max	Min	Std error
Benzene	ppb	0.487	0.000	1.100	0.194
Ethylbenzene	ppb	0.000	0.000	0.000	0.000
Toluene	ppb	0.390	0.000	0.870	0.148
Xylenes	ppb	0.000	0.000	0.000	0.000
Methane	ppm	2.700	2.300	3.200	0.144
PM 10	mg/m3	0.062	0.013	0.107	0.018
VOC	ppb	46.42	1.000	153.5	24.57
Flowback	Units	Average	Max	Min	Std error
Benzene	ppb	16.29	0.230	82.00	13.30
Ethylbenzene	ppb	1.583	0.000	7.900	1.290
Toluene	ppb	14.76	0.180	73.00	11.82
Xylenes	ppb	4.588	0.000	22.00	3.560
Methane	ppm	15.50	10.00	32.00	4.491
PM 10	mg/m3	0.004	0.001	0.012	0.002
VOC	ppb	853.6	333.5	1377	226.9
Production	Units	Average	Max	Min	Std error
Benzene	ppb	0.400	0.000	1.200	0.400
Ethylbenzene	ppb	0.000	0.000	0.000	0.000
Toluene	ppb	0.983	0.000	2.600	0.815
Xylenes	ppb	0.000	0.000	0.000	0.000
Methane	ppm	3.500	3.200	3.700	0.153
PM 10	mg/m3	1.777	0.010	5.230	1.727
VOC	ppb	7.667	3.500	10.00	2.088
Midland	Units	Average	Max	Min	Std error
Benzene	ppb	0.327	0.000	0.540	0.166
Ethylbenzene	ppb	0.000	0.000	0.000	0.000
Toluene	ppb	0.860	0.640	1.000	0.111
Xylenes	ppb	0.000	0.000	0.000	0.000
Methane	ppm	3.100	2.700	3.300	0.200
PM 10	mg/m3	0.060	0.023	0.097	0.021
VOC	ppb	25.17	65.00	2.500	19.98

Nearly all air-monitoring parameters measured during fracturing and in the production phases were lower or comparable to the HFTS baseline values, the Midland baseline, and literature values for the DFW and Fort Worth measurements (Rich, et al., 2011; Bunch, et al., 2014). PM-10 values were not detected in the high quantities expected during hydraulic fracturing from to truck traffic and sand loading. HFTS operations used high quality filters on all silica proppant silos to prevent particulate matter pollution, and the installation and use of water, gas, and oil pipelines across the Laredo facility limited truck traffic during this time period. The highest 24-hour average of PM 10 was measured during production, at 5.23mg/m^3, OSHA acceptable levels for 8-hour time weighted average is 15 mg/m^3.

Median and average concentrations of BTEX, total VOCs, and methane are displayed in the bar charts of **Figure 3**. Average values for BTEX, VOCs and methane for the hydraulic fracturing and production phases were either less than or comparable to the HFTS Baseline and Midland air monitoring values. BTEX, VOC and methane concentrations during the flowback phase were substantially greater in magnitude in terms of average and median values than the air monitoring results obtained from the baseline, fracturing, and production phases, as well as from the Midland location. This difference was not significant when analyzed with ANOVA for BTEX (p-value=0.18), VOC (p-value=0.92), or methane (p-value=0.52).



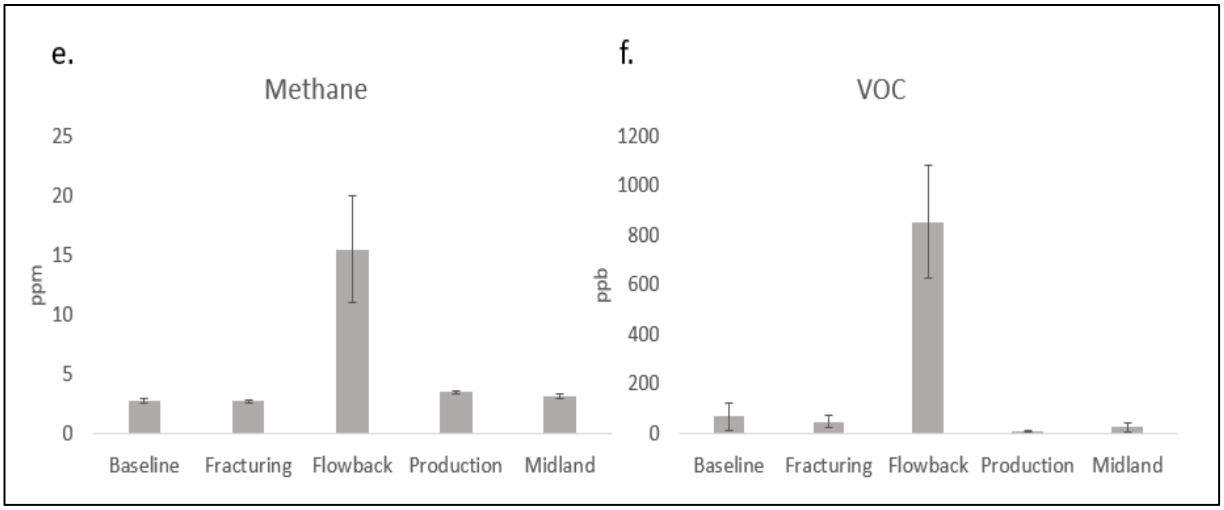


Figure 3: BTEX, Methane, and VOC 12 hour Averages during HFTS Experiment

Overall, our limited air monitoring effort indicates the HFTS experiment had a minimal impact on concentrations of key pollutants related to oil and gas development. The concentrations of BTEX, Methane, and VOCs 1,000 ft. from the well pad increased during the flowback period. The concentrations of BTEX measured at HFTS were orders of magnitude lower than the OSHA limits for worker safety exposure. The authors acknowledge the improper comparison between our 12-hour average data and OSHA's 8-hour averages, however, the merit of the compassion is relevant as the highest recorded 12 hour HFTS concentrations of benzene (82 ppb) are several orders of magnitude below OSHA's 8-hour limits (1,000 ppb). If mitigation is deemed necessary, granular sorbents (such as granular activated carbon) would likely be effective and economical in the control of BTEX and VOCs emitted from frac tank vents.

Water Characterization

Characterization data on the composition of the impoundment water used for hydraulic fracturing are shown in **Table 4**. Impoundment water includes two freshwater impoundments and one recycled produced water impoundment known as the Poseidon tank. Fracture fluids were a mixture of fresh and recycled produced water (80/20), samples were obtained after chemical mixing before proppant addition and pressurization at the first, sixth, and final day of hydraulic fracturing. Hydraulic fracturing of each well occurred over 12-13 days. All impoundments and the fracture fluids themselves were of normal alkalinities at pH levels ranging 6.2 – 8.3. Impoundment water had total organic carbon (TOC) concentrations of about 1-2 mg/L, fracture fluids around 150 mg/L. The salinity of the freshwater impoundments showed a modest level around 2,000 to 5,000 mg/L of TDS, while the water from the Poseidon storage unit exhibited more than 20 times that ionic strength (75,000 to over 100,000 mg/L); salinity of all the impoundments was comprised mostly of sodium and chloride.

Table 4: Characteristics of Impoundments that Store Water for Hydraulic Fracturing at the HFTS.

	Freshwater Impoundment*		Poseidon Impoundment**		Fracture Fluids***	
Parameter	Range	Median	Range	Median	Range	Median
pH	8.03-8.3	8.17	6.64-7.03	6.81	6.28-7.08	6.66
TDS, mg/L	1,790-3,800	2,580	75,400-104,000	98,500	3,370-25,300	21,700
Conductivity, µS/cm	1,460-5,270	3,790	13,000-69,100	47,850	4,960-29,500	18,200
Alkalinity, mg/L as $CaCO_3$	143-183	177	236-383	344	114-190	178
Total Organic Carbon, mg/L	1.97-5.4	2.2	0.4-1.1	0.98	87-180	150

Chloride, mg/L	149-1,380	793	730-1,380	1,220	1,310-13,800	12,200
Nitrate, mg/L	6.7-15.1	13	<1	<1	2-11	8
Sulfate, mg/L	340-650	494	185-310	230	342-570	490

* Freshwater Impoundment: Six samples obtained and analyzed per impoundment (2).
** Treated Produced Water (Saline Water) Impoundment: Six samples taken and analyzed.
*** Fracture Fluids: Fracture fluids after mixing and chemicals added to Impoundment water. Three samples per well were taken and analyzed.

Sulfate which has a high potential of biotransformation is present in all of the impoundment and fracture fluids at substantial concentrations. Sulfate can be converted to sulfide (e.g. H_2S) through the action of sulfate reducing bacteria. Sulfate in freshwater impoundments was present at around 500 mg/L, while the recycled produced water impoundment had a median value of around 230 mg/L; a 80/20 mixture of freshwater to saline would result in sulfate levels of over 400 mg/L in streams used for the hydraulic fracturing operations. With the introduction of 10 million gallons of fracture fluids containing 400 mg/L of sulfate, it is likely that more than 16.5 tons of sulfate was introduced into the subsurface during the completion of each shale well; at this level of mass input of sulfur to the subsurface, it becomes important to understand the degree to which sulfate is involved in biological processes that result in accelerated corrosion (i.e. microbial influenced corrosion or MIC), the microbial production of sour gas (i.e. H_2S), or the deposition of iron sulfides. Another compound with potential for biotransformation is nitrate which can be converted to nitrogen through the action of denitrifying bacteria. Nitrate levels in the freshwater impoundments showed median values of over 12 mg/L, though even higher levels of nitrate could be possible if completion operations involved the use of nitric acid for cleaning.

Quantitative PCR of the 16S rRNA gene (an estimation of total Bacteria (BA)) in surface impoundments and fracture fluids revealed fresh water bacterial concentration to be around $1x10^4$ 16S rRNA gene copies per mL in fresh water impoundments and concentrations up to $1x10^8$ 16S rRNA gene copies per mL of water in the Poseidon impoundment. The differences in BA concentration are expected as Poseidon water is recycled produced water that is stored in open top pits without anti-microbial treatment. Drill out muds, used for drilling out the fracture plugs, also contained a high concentration of bacteria, averaging $1x10^7$ copies per mL. Bacteria ranged in concentration from $6x10^4$ to $5x10^7$ 16S gene copies per mL in fracture fluids over the course of 12-14 days of hydraulic fracturing. This moderate to high concentration of bacteria introduced to each well during hydraulic fracturing has been documented in other cases (Struchtemeyer, 2011) and is recognized as a significant source of potentially metabolically active microorganisms to the subsurface.

Upon initiation of flowback approximately six weeks after hydraulic fracturing, substantial increases in salinity are documented in the produced water (**Figure 4**). Also indicated in this graph is the level of average TDS levels measured in produced water obtained from three mature shale wells operating in the Wolfcamp formation for three years with an average of 124,000 mg/L TDS; this was done to show the levels of salinity that are reached as a result of long term saturation of downhole minerals and mixing with formation water.

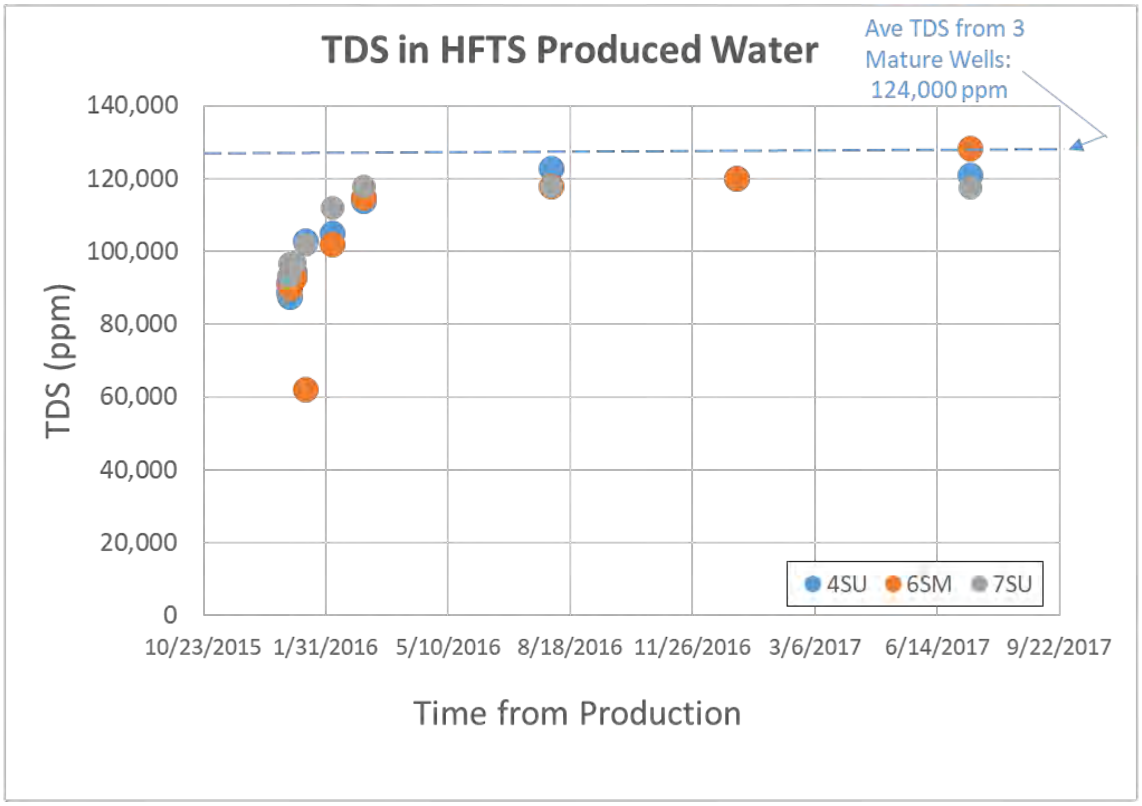


Figure 4: Total Dissolved Solids with Time in Produced Waters Collected from the Three Shale Wells at the HFTS

At many shale wells monitored in the Marcellus and in the Barnett plays, profiles of salinity measurements in flowback waters that are released within a few days following hydraulic fracturing show an initial TDS level that is less than 20% of the ultimate saturation concentration (Hayes, 2009; Hayes and Severin, 2012). The profiles of the monitored wells of HFTS, however, were quite different; the plot of **Figure 4** shows that the initial flowback water stream had TDS levels of about 90,000 mg/L TDS, more than 75% of the ultimate saturation level of 120,000 mg/L. This was likely due to the relatively longer contact time (6 weeks) that the flowback water had with formation mineral materials that allowed the water to come closer to salt saturation before the initiation of flowback water release.

As modest increases in salts were observed in the flowback and produced water collected from each of the three sampled shale wells, decreasing concentrations in other constituents were noted that indicated possible dilution and transformational actions were taking place in the downhole environment. Plots of sulfate, nitrate and total organic carbon (TOC) with time following the initiation of release of flowback/produced water are shown in **Figures 5, 6 and 7,** respectively. Over the initial 14-day interval, sulfate concentrations were decreased from approximately 422 mg/L down to 318 mg/L, a 104 mg/L reduction (Figure 5). In the same period, nitrate concentrations fell from 36 mg/L to 0 mg/L (Figure 6). Simultaneously, soluble organics measured as total organic carbon (TOC) showed a decrease of approximately 36 mg/L in the flowback during the first 14 days as seen in Figure 7.

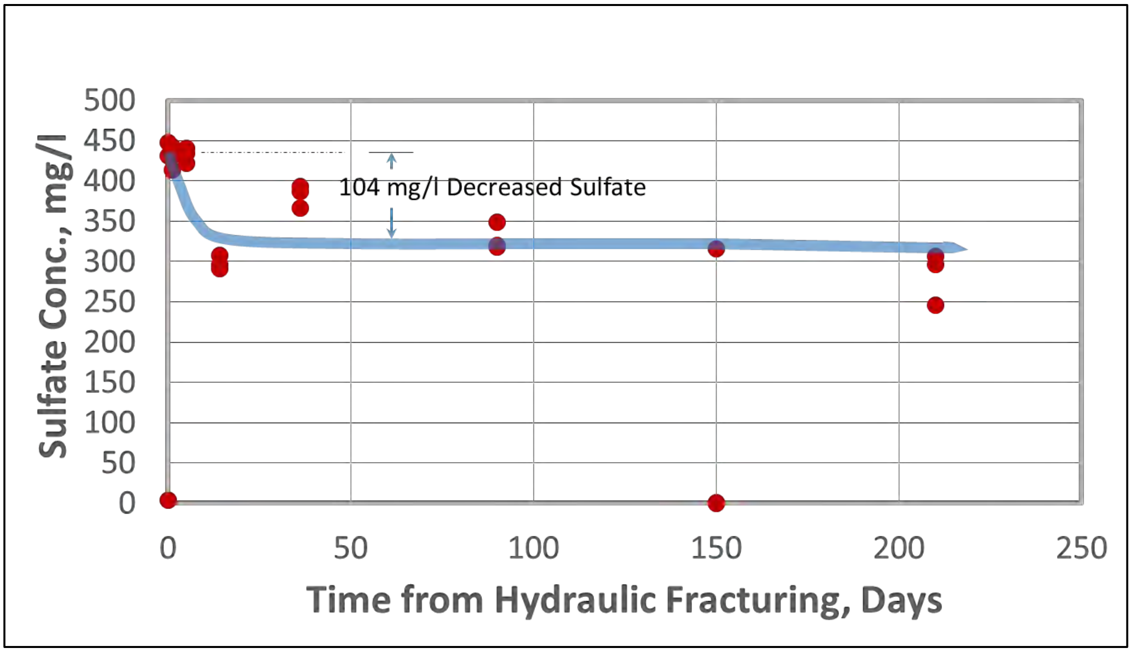


Figure 5: Sulfate Vs. Time in Produced Waters Collected from Three Wells Selected for Sampling at the HFTS

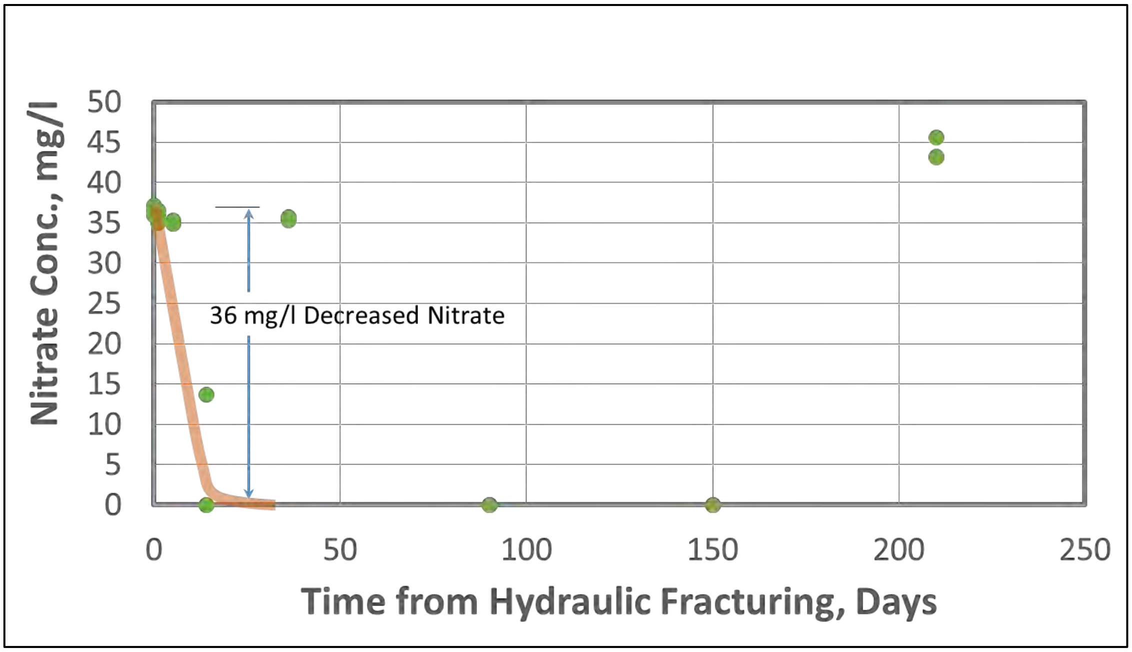


Figure 6: Nitrate Vs. Time in Produced Waters Collected from the Three Wells Selected for Sampling at the HFTS

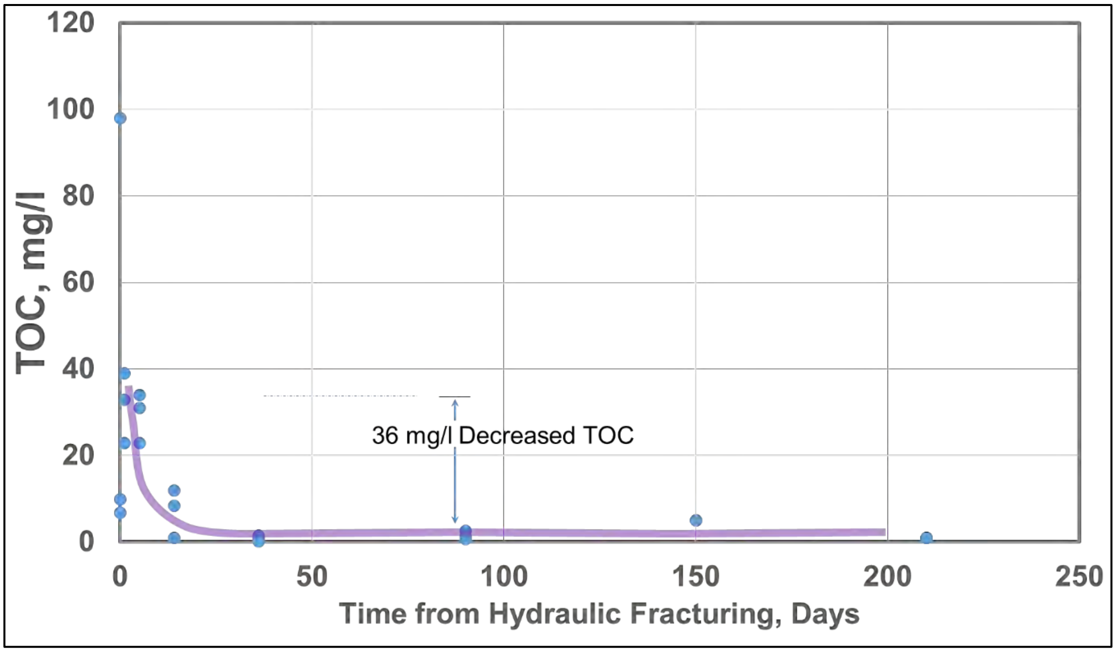


Figure 7: Total Organic Carbon Vs. Time in Produced Waters Collected from Three Wells at the HFTS

These observed changes suggest the occurrence of microbial-mediated transformations where the oxygen content of nitrate and sulfate is used to convert organic compounds to carbon dioxide and subsequently to methane. One group of microorganisms, the denitrifying bacteria, are able to utilize nitrate to oxidize organic compounds to carbon dioxide while reducing nitrate to nitrogen gas (Brock, 1970). Another group of microbes, sulfate reducing bacteria (SRBs), are able to utilize sulfate to oxidize organic compounds to carbon dioxide while reducing sulfate to hydrogen sulfide (Stumm and Morgan, 1981). Methanogens present in the formation are then able to generate methane from the microbiologically produced carbon dioxide.

Average bacteria concentration, as measured by qPCR in all three wells over the first year and a half of production is shown in **Table 5** along with sulfate-reducing bacteria (SRB), denitrifying bacteria (DNB), and methanogen (Meth) concentrations. Total bacterial populations peaked at 1.3x10^8 16S genes per mL after 36 days of production in well 6SM, the lowest measured concentration of bacteria to date is 1x10^4 16S genes per ml after 210 days of production in well 4SU. Concentrations of SRB increased over the first year and a half of production and concentrations of DNB decreased in time with the decline in nitrate concentrations. Methanogens were detected in moderate concentration throughout the lifetime of the well. The deleterious effect of microbiological activity was witnessed within three months of production, when unplanned downtime was necessary to replace tubing in one of the wells due to extensive microbially induced corrosion (MIC). Within 6 months of production, biocide treatment was necessary in all 11 wells.

Table 5: Average Copies / mL of Bacteria (BA), Sulfate Reducing Bacteria (SRB), Denitrifying Bacteria (DNB), and Methanogens (Meth) over 550 of Production at the HFTS

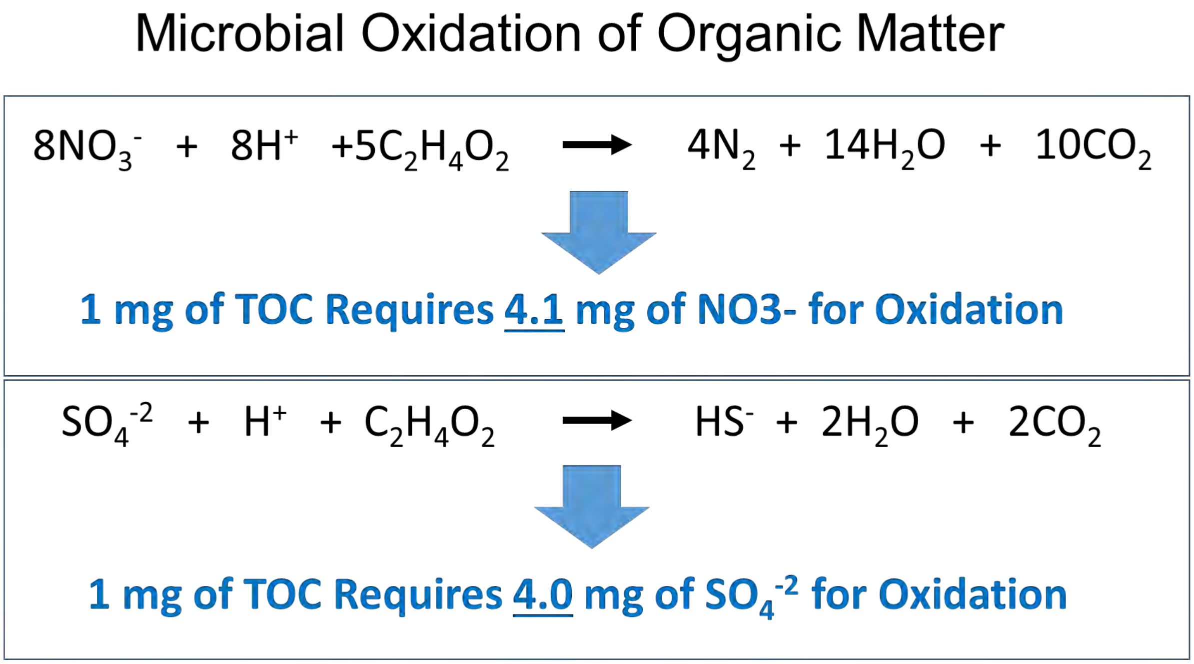
		Average copies / mL in Produced Water			
ID	**Date**	**BA**	**SRB**	**DNB**	**Meth**
PW0	**12/31/2015**	1.13E+05	1.29E+01	3.31E+02	2.70E+04
PW1	**1/1/2016**	3.25E+07	3.91E+01	7.58E+02	3.08E+03
PW6	**1/5/2016**	6.96E+05	3.73E+02	1.78E+01	7.69E+03
PW14	**1/14/2016**	1.46E+07	1.12E+02	1.37E+02	3.76E+03
PW36	**2/5/2016**	5.34E+07	1.73E+03	4.95E+01	2.31E+04
PW90	**3/2/2016**	1.56E+07	9.62E+02	4.09E+00	2.89E+03

PW210	**8/2/2016**	6.57E+04	4.54E+02	3.70E+00	6.34E+02
PW365	**12/30/2016**	9.51E+05	8.14E+03	2.31E+00	8.24E+03
PW550	**6/30/2017**	3.41E+06	1.68E+03	9.61E+00	4.31E+03

Metagenomic sequence (16S rRNA gene) analysis revealed consistent, increasing populations of sulfur cycling microorganisms including *Arcobacter,* which form filamentous sulfur mats (Wirsen, 2004) aiding in bacterial attachment to rocky surfaces with high fluid flow. Also found were *Archaeoglobus*, which are sulfate reducing archaea with optimal growth at 83°C, commonly found in oilfield systems with the ability to reduce sulfate to sulfide with the oxidation of complex carbon polymers, several types of sulfate-reducing bacteria including *Desulfonatronum, Desulfovibrio, Desulfomondaceae, Desulfonatronovibrio, Desulfovermiculus, Desulfobacteraceae*, and a large persistent population of *Halanaerobium*. *Halanaerobium* are fermentative bacteria often found in produced water, which feed on ethylene glycol, a common additive to fracture fluids. These bacteria produce hydrogen, which is then a primary source of energy for the SRB and Methanogens. *Halanaerobium* also reduce thiosulfate to hydrogen sulfide. The microbiological data and observed microbiological action of MIC support the theory of microbiological transformation of sulfate in the stimulated reservoir volume (SRV).

When organic matter is not available for conversion by microbial communities, both sulfate and nitrate are stable and persist in the subsurface aquatic environment, as evidenced by groundwater composition data in many areas of Texas as well as in the baseline groundwater composition data for the five wells adjacent to the HFTS facility. In the case of the groundwater wells adjacent to the HFTS location, **Table 2** shows values for sulfate and nitrate at about 65-390 mg/L and 0.1-25 mg/L, respectively, which persist at these levels in the aquifer environment; this is due to the low activity of key microbial communities in the aquifer (i.e. denitrifiers and SRBs) and the very limited availability (i.e. low concentration) of total organic carbon (at 2 mg/L or less). When the same water is blended with chemicals and injected downhole during hydraulic fracturing, the soluble organic carbon rises significantly to levels that may possibly stimulate introduced or *in-situ* microbial communities to utilize sulfate and nitrate to convert (oxidize) some or all of the soluble carbon to produce carbon dioxide.

If the availability of soluble organic carbon is key to the microbiological reduction of nitrate and sulfate, then the oxygen equivalent of the TOC consumed should be roughly matched to the oxygen equivalent of the nitrate and sulfate that was remove from solution during the same period. Balanced equations that describe denitrification and sulfur reduction using organic matter are shown in **Figure 8**, using acetic acid as the example of organic matter that is oxidized. Between the two types of oxidation that occur (denitrifying and sulfur reducing), one milligram of TOC requires roughly four milligrams of nitrate or sulfate to achieve complete conversion of the organic compound to CO_2.



Microbial Oxidation of Organic Matter

$8NO_3^- + 8H^+ + 5C_2H_4O_2 \rightarrow 4N_2 + 14H_2O + 10CO_2$

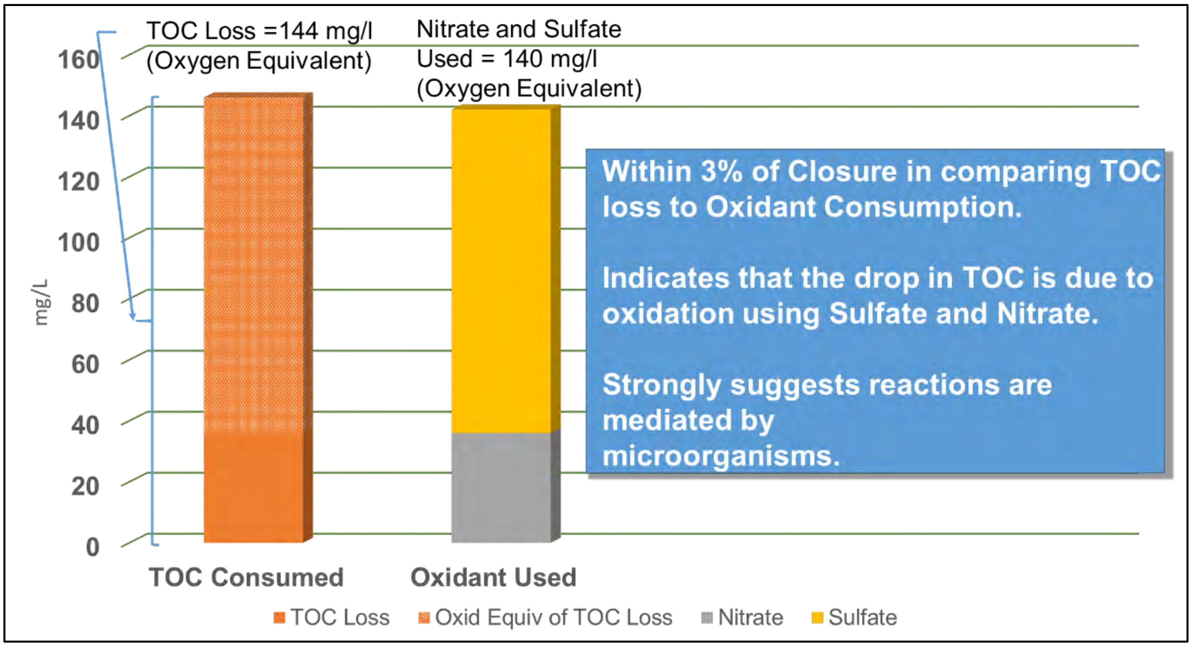
1 mg of TOC Requires 4.1 mg of NO3- for Oxidation

$SO_4^{-2} + H^+ + C_2H_4O_2 \rightarrow HS^- + 2H_2O + 2CO_2$

1 mg of TOC Requires 4.0 mg of SO_4^{-2} for Oxidation

Figure 8: Balanced Equations Describing the Microbial Oxidation of Organic Matter Using Nitrate and Sulfate

When these stoichiometric conversion factors are considered along with the concentration reductions during the first 14 days of flowback release, it appears that the decreased concentration of TOC in the downhole water in terms of oxygen equivalent is close in value to the oxygen equivalent of sulfate and nitrate consumed during this same period. This is shown in a comparison of oxidant and TOC conversion shown in **Figure 9** where the loss of soluble TOC (oxygen equivalent) came within 3% of the consumption of nitrate and sulfate in terms oxygen equivalents. This would indicate that the drop in soluble TOC is due to oxidation using nitrate and sulfate. More important, this would strongly suggest that the oxidative transformations of TOC utilizing sulfate and nitrate are mediated by microbial consortia in the downhole environment.



Within 3% of Closure in comparing TOC loss to Oxidant Consumption.

Indicates that the drop in TOC is due to oxidation using Sulfate and Nitrate.

Strongly suggests reactions are mediated by microorganisms.

Figure 9: Comparison of Oxidant and TOC Conversion in the Downhole Aquatic Environment During the Initial 14 Days of Flowback Release

If the concentrations in the produced water samples are indicative of the levels of TOC, sulfate and nitrate in the SRV (with a total initial volume of 10 million gallons per completed well), then mass flow calculations show that within two weeks following the release of flowback about 1.5 tons of TOC was converted to CO_2 through the action of microbes with the concomitant reduction of 1.5 tons of nitrate to nitrogen gas and 4.2 tons of sulfate to sulfide amounting to about 1.4 tons. While the nitrogen gas is inert, the formation of over a ton of sulfide from sulfate conversion could potentially be problematic in terms of corrosion, deposition, or sour gas formation. If all of the 104 mg/L of transformed sulfate was converted to soluble sulfide and distributed in the formation, the resultant concentration of soluble sulfide would be at least 30 mg/L as HS^-.

The results of sulfide analysis of all flowback and produced water samples, however, show soluble sulfide levels at below detection (< 1 mg/L). This is likely due to the presence of soluble iron at concentrations of 15 to 170 mg/L in the downhole aquatic environment; at these concentrations, iron levels at the pH level of the produced water (pH 7.5 to 8.0) may be sufficient to precipitate the sulfide as ferrous sulfide, thereby reducing soluble sulfide concentrations to below 1 mg/L. A similar observation of low sulfide (<1.2 mg/L) in the presence of 25-200 mg/L of iron has also been described in another study where produced waters were characterized at 19 locations in the Marcellus Shale (Hayes, 2010) and in the Barnett Shale (Hayes and Severin, 2012). Sulfide may also be complexed with metals associated with the well corrosion materials and be rapidly generated in biofilms involved with microbiologically influenced corrosion experienced in all 11 wells during the HFTS experiment.

Alternative transformations which could occur in the subsurface, attributing to a decrease in measurable sulfate include the formation of calcium sulfate or barium sulfate scale. However, levels of Ca in produced water increased from 1,900ppm to 2,410ppm over the year and a half of production. Ba concentration was low, between non-detect and 2.5mg/L over the year and a half of production. Therefore, we do not have strong evidence of Ca or Ba reacting with sulfate to create scale at a large enough scale to lead to the marked decline in sulfate in PW over time.

Microbial Control

Biocides are used in the oil and gas industry to prevent the breakdown of fracture fluid chemicals, microbiologically induced corrosion, sour gas production, and decreased formation permeability due to biofilm. Our microbiology results indicated high concentrations of bacterial populations were introduced into the wells and persisted in the subsurface, up to 550 days after production. The detrimental impact of biofilms was observed on site as corrosion failures, and potentially through decreased production rates due to microbiological transformation of sulfate to iron sulfide deposits. Biocides were used at HFTS twice: once to pre-treat fluids used for hydraulic fracturing and six months into production to mitigate the impact of corrosive microbiological activity on downhole equipment including tubing and gauges.

Two factors likely contributed to the establishment of viable microbial communities in the subsurface. First, recycled produced water used for hydraulic fracturing was not treated to adequately kill all microorganisms before hydraulic fracturing, as communicated by Laredo operations. Recycled produced water at the HFTS was treated with chlorine dioxide, a strong oxidizing biocide, and stored in impoundment tanks before being mixed with fresh water, fracturing fluid chemicals and sand, and then injected into the reservoir. This introduced living, thermo- and halo-tolerant, biofilm promoting organisms into the SRV. Secondly, the wells were shut in for a period of six weeks after hydraulic fracturing. Post-fracture biocide treatments are found to be most successful when flowback occurs within 24 hours of hydraulic fracturing (Struchtemeyer C. 2012). The shorter residence time of fracturing fluids limits the amount of time for cell – surface interactions required for anaerobic biofilm growth and proliferation. Once established however, biofilms are notoriously difficult to remove from the formation and steel surfaces, so produced water microbiology must be monitored and managed.

In November and early December 2016, wells were taken offline and shut in for 12 hours for biocide treatment after microbiologically influenced corrosion was observed on tubing and pressure gauges. Approximately 55 gallons of B-1203, a Glutaraldehyde/ Didecyldimethylammonium Chloride blend, was pumped downhole. Gluteraldehyde binds to cell's amine groups, killing microorganisms by inhibiting the transport of essential nutrients. Didecyldimethylammonium chloride, is a quaternary ammonium compound, otherwise known as "quats". Quats are added to glutaraldehyde biocides for their ability to disrupt cell membranes and grant access for amine binding. Quaternary compounds are most effective on Gram positive cell membrane structure. Sequencing results of produced water revealed the majority or organisms growing in the reservoir since the early days of production were Gram negative.

The qPCR results from samples taken one month after biocide application indicate bacteria concentrations have remained steady or increased in some wells (**Table 5**). The persistence of high concentrations of bacterial DNA in the subsurface indicates an incomplete kill job in these wells. Although live vs. dead tests were not conducted for these samples, consistent and increased population concentrations sampled over time with quantitative methods indicates populations are alive and sustaining growth in the SRV. Furthermore, the sequencing of DNA in samples taken days after the biocide treatment indicate the subsurface population is now dominated by Gram negative organisms, and organisms with natural defenses to quats such as double membranes or protective sheaths around the cell body. There are strategies for enhancing biocide effectiveness against Gram negative bacteria (Denyer 2002), which can be determined based on fracturing fluid and formation chemistry.

Incomplete kill jobs from biocide treatments is common in the unconventional oil and gas industry (Gaspar J. 2014). Chemically, quaternary ammonium compounds work best in soft water, with calcium concentrations below 60 mg/L. The ground water introduced to the reservoir during hydraulic fracturing at HFTS was only moderately hard (Ca = 75 mg/L), however the produced water has Ca >2,000 mg/L. The biocide treatment in November introduced 55 gallons of B-1203 to a reservoir containing considerably hard water, likely compromising the biocide effectiveness. Microbial response to biocide choice and duration for fracturing fluids to be in contact with the formation, indicate operation activities do have an influence on ultimate subsurface microbiological community structure and activity.

Conclusions

In consideration of the monitoring results obtained by this evaluation effort, the following summary conclusions are drawn:

1. Application and testing of advanced technologies of drilling, completion and production at the HFTS facility did not result in lasting deterioration of groundwater. In the monitoring of groundwater before and during each phase of operation, no evidence of migration of methane, benzene, toluene, ethylbenzene, xylenes or total organic carbon (TOC) was detected.

2. The temporary rise in median salinity of the groundwater measured at the five groundwater wells (increasing from about 500 mg/l to 800 mg/l) was likely due to the infiltration of water from deeper portions of the aquifer of higher salinity during the fracturing phase when extensive pumping (amounting to tens of millions of gallons) could have caused groundwater drawdown of each of the five wells. The effect was short termed: dissolved solids returned to baseline TDS levels within 50 days.

3. Air monitoring measurements conducted at the HFTS including VOC, PM-10, benzene, toluene, ethylbenzene, total xylenes, and methane indicate that nearly all phases of HFTS operations (with the exception of flowback collection) had no discernable impact to air quality when compared to the baseline HFTS samples, monitoring conducted in downtown Midland, and to literature values from other urban locations in Texas. It is recognized that there are some limitations to the sampling protocol.

4. The collection and storage of initial produced water released from each well (flowback) does result in measurable release of VOCs and BTEX to the atmosphere; however, near-site air quality values for BTEX are less than 10% of OSHA limits for worker safety (8-hour exposure) and the short-term nature of the flowback collection phase limits local residential exposure to air emissions. If needed, however, these small emissions can be economically controlled through engineering controls (adsorbents) applied to frac tank vents.

5. Delaying flowback release by 6 weeks following the completion of each well provided contact time for the injected fluids to equilibrate with downhole materials and causes higher salinities to be measured in early flowback samples. Upon release, median TDS levels in flowback water were measured at around 90,000 mg/l in the first day of release increasing to a plateau of 120,000 mg/l in about 3 months, close in value with the salinity of produced waters generated from mature wells of nearby Wolfcamp shale well fields.

6. As salinity in the flowback increased with time, concentrations of several constituents decreased simultaneously, the nature of which strongly pointed to the likelihood of substantial microbial mediated transformations of hydrocarbons and oxidants. Within the first 14 days following initial release of flowback, TOC decreased by 95% while nitrate decreased by 100% (mediated by denitrifier microbial populations) and sulfate (initially present at over 420 mg/l) decreased by 24% (mediated through the action of sulfate reducing bacteria). The measured TOC loss was in stoichiometric balance with nitrate and sulfate consumption: The oxygen equivalents of TOC oxidized to CO2 were within 3% of the measured losses of sulfate and nitrate oxygen equivalents in the flowback at 14 days. Chemical data is supported by quantitative microbiological analysis of denitrifier and sulfate reducing bacteria.

7. If the changes in concentration in the flowback water samples were indicative of the 10 million gallons of injected water, more than 1.4 tons of sulfide could have been generated in place in the fractured shale. The non-detect levels of soluble sulfide measured in the flowback water samples together with the presence of excess soluble iron point to the substantial formation of iron sulfide precipitate. More than 3.8 tons of ferrous sulfide precipitate may have formed along the lateral of each horizontal well. This in-place precipitate may later move toward the horizontal well bore as water is removed and may over time mechanically impede the movement of fluids and gases from a horizontal well and become a factor in overall well performance. More research will be needed to determine the fate of sulfide and management approaches to prevent deleterious effects on well performance.

8. All 11 wells experienced microbiologically influenced corrosion within six months of production. Subsequent biocide application was not adequate to limit microbial growth and activity. Biocide dosing decreased the population of methane producing bacteria, however it did not substantially reduce overall bacterial populations sizes or the relative abundance of sulfate reducing bacteria. The biocide used targeted Gram positive bacteria and works optimally in soft water (<60mg/L Ca). At the time of biocide treatment bacterial populations were composed of primarily Gram negative organisms with a Ca concentration over 2,000 mg/L.

References

Anaya, R. and I. Jones. 2009. “Groundwater Availability Model for the Edwards-Trinity (Plateau) and Pecos Valley Aquifers of Texas: Texas Water Development Board Report 373. (103 pages).

Brock, T. 1970. Biology of Microorganisms. Prentice Hall, Inc. Englewood, NJ. pp. 486-487.

Bunch, A.G., et al. 2014. “Evaluation of Impact of Shale Gas Operations in the Barnett Regions on Volatile Organic Compounds and Potential Health Risks.” Science of the Total Environment: 468-469. pp. 832-842.

Clark, C., A. Burnham, C. Harto, and R. Horner. 2013. “Hydraulic Fracturing and Shale Gas Production: Technology, Impacts, and Regulations.” USDOE Office of Scientific and Technology Information, Oak Ridge, TN. Report No. ANL/EVS/R-12/5.

Daly, Borton, Wilkins, Hoyt, Kountz, Wolfe, Welch, Marcus, Trexler, MacRae, Krzychki, Cole, Mouser, Wrighton. 2016. "Microbial metabolisms in a 2.5km deep ecosystem creasted by hydraulic fracturing in shales." *Nature Microbiology* 16146.

EIA. 2017. “International Energy Outlook 2017.” Report No. IEO2017. U.S. Energy Information Administration. p. 55.

Eisenlord, S. and T. Hayes. 2017. “Microbiological Analysis of Hydraulic Fracture Test Site (HFTS) Operations. Final Report. Gas Technology Institute, Des Plaines, IL.

Galperin, Kaplan. 2011. "Review of microbial processes in the near-surface environment and their implications for the chemical fingerprinting of hydrocarbon fuels." *Environmental FOrensics* 236-252.

Gaspar J., J Mathiew, Y Yang, R Tomson, J Leyris, K Gregory, P Alvarez. 2014. "Microbioal dynamics and control in shale gas production." *Enviornmental Science and Technology* 465-473.

Gallegos, T.J., B.A. Valera, S.S. Haines, and M.A. Engle. 2015. Hydraulic Fracturing in the U.S.” AGU Water Resources Research. pp 5830-5845.

Hayes, T.D. 2010. “Sampling and Analysis of Water Streams Associated with the Development of Marcellus Shale Gas,” Final Report to the Marcellus Shale Coalition. Gas Technology Institute, Des Plaines, IL.

Hayes, T.D. and B.F. Severin. 2012. “Barnett and Appalachian Shale Water Management and Reuse Technologies,” RPSEA Report No. 08122-05.FINAL.2.

Hayes, T.D. and B.F. Severin. 2012. “Characterization of Flowback Waters from the Marcellus and the Barnett Regions.” RPSEA Report No. 08122-05.09.

Lipus, Vikram, Ross, Bain, Gulliver, Hammack, Bibby. 2017. "Prdominance and metabolic potential for halanaerobium in produced water from hydraulically fractures marcellus shale wells." *Applied and Environmental Microbiology* Online.

Magot, Olliver, Patel. 2000. "Microbiology of petroium reservoirs." *Antonie van Leeuwenhoek* 103-116.

Mohan, Harstock, Hammack, Vidic, Gregory. 2013. "Microbial communities in flowback water impoundments from hydraculic fracturing for recovery of shale gas." *FEMS Microbiology Ecology* 1-14

Rich, A., J.P. Grover, and M.L. Sattler. 2014. "An Exploratory Study of Air Emissions Associated with Shale Gas Development and Production in the Barnett Shale." Journal of the Air and Waste Management Association. Vol 64(1): 61-72.

Roling, Head, Larter. 2003. "The microbiology of hydrocarbon degradation in subsurface petroleum reservoirs: perspectives and prospects." *Research in Microbiology* 321-328Small, M.J., et al. 2014. "Risk and Risk Governance in Unconventional Shale Gas Development." ES&T, 48: 8289-8297.

Struchtemeyer C., M Elshahed. 2011. "Bacterial communities associated with hydraulic fracturing fluids in thermogenic natural gas wells in North Central Texas, USA." *FEMS Microbial Ecology* 13-25.

Strutchtemeryer, Morrison, Elshaed. 2012. "A crittical assessment of the efficacy of biocides used during the hydraulic fracturing process in shale natural gas wells." *International Biodeterioration & Biodegradation* 15-21.

Stumm, W. and J.J. Morgan. 1981. Aquatic Chemistry. John Wiley & Sons, Inc., Hoboken, NJ. pp. 458-461.

USEPA. 2016. "Hydraulic Fracturing for Oil and Gas: Impacts from the Hydraulic Fracturing Water Cycle on Drinking Water Resources in the U.S." Report No. EPA-600-R-16-236Fa. December 2016.

Voordouw, G., Grigoryan, Lambo, Lin, Park, Jack, Coombe, Clay, Zhang, Ertmoed, Miner, and Arensdorf. 2009. "Sulfide remediation by pulsed injection of nitrate into a low temperature candian havy oil reservoir." *Environemtnal Science and Technology* 9512-9518.