

DISSERTATION

THE DOWNHOLE BEHAVIOR OF THE CHEMICALS OF HYDRAULIC FRACTURING –
AN INSIGHT TO THE NATURE OF BIOCIDES AND SURFACTANTS UNDERGROUND

Submitted by

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ABSTRACT

THE DOWNHOLE BEHAVIOR OF THE CHEMICALS OF HYDRAULIC FRACTURING – AN INSIGHT TO THE NATURE OF BIOCIDES AND SURFACTANTS UNDERGROUND

In a time period and society surrounded by a surplus of information, there is currently mystery and confusion surrounding the organic chemicals added to hydraulic fracturing (“fracking”) fluids. Not only is it unclear what chemicals specifically are being used in some instances, but there is little to no information existing about the transformations these chemicals may undergo once underground (“downhole”) and subjected to elevated heat and pressure for the duration of a fracturing operation. Several kilometers downhole, these organic chemicals are exposed to temperatures up to 200 °C, pressures above 10 MPa, high salinities, and a pH range from 5 - 8. Despite this, very little is known about the fate of HFF additives under these extreme conditions. Chemical transformations may directly affect the toxicity of the chemicals as they emerge from the downhole environment with the rest of the “flowback” wastewater. Therefore the following chapters of this dissertation serve to classify existing information and to probe the basic effects of the downhole fracturing environment on chemical stability and transformation.

Chapter 1 provides a brief introduction to and rationale for the research presented in the following pages. Some of the general purposes for chemicals within hydraulic fracturing fluids (HFFs) are discussed, as well as some of the reason for the controversy which exists today. Additionally, chapter 1 outlines the research objectives which inspired the original research presented afterwards.

Chapter 2 of the dissertation serves as the first existing literature review on the biocides utilized in hydraulic fracturing. Biocides are critical components of hydraulic fracturing (“fracking”) fluids used for unconventional shale gas development. Bacteria may cause bioclogging and inhibit gas extraction, produce toxic hydrogen sulfide, and induce corrosion leading to downhole equipment failure. The use of biocides has spurred a public concern and debate among regulators regarding the impact of inadvertent releases into the environment on ecosystem and human health. Chapter 2 provides a review of the potential fate and toxicity of biocides used in hydraulic fracturing operations. Physicochemical and toxicological aspects will be discussed as well as knowledge gaps that should be considered when selecting biocides: (1) uncharged species will dominate in the aqueous phase and be subject to degradation and transport whereas charged species will sorb to soils and be less bioavailable; (2) many biocides are short-lived or degradable through abiotic and biotic processes but some may transform into more toxic or persistent compounds; (3) understanding of biocides’ fate under downhole conditions (high pressure, temperature, salt and organic matter concentrations) is limited; (4) several biocidal alternatives exist, but high cost, high energy demands, and/or formation of disinfection byproducts limit their use.

Chapter 3 serves as the first research experiment outlining a model for testing the behavior of HFF additives downhole. Here, stainless steel reactors are used to simulate the downhole chemistry of the commonly used HFF biocide glutaraldehyde (GA). The results show that GA rapidly ($t_{1/2} < 1$ hr) autopolymerizes, forming water-soluble dimers and trimers, and eventually precipitates out at high temperatures (~ 140 °C) and/or alkaline pH. Interestingly, salinity was found to significantly inhibit GA transformation. Pressure and shale did not affect GA transformation and/or removal from the bulk fluid. Based on experimental second-order rate

constants, this chapter provides a working kinetic model for GA downhole half-life predictions for any combination of these conditions (within the limits researched) was developed. The findings outlined in chapter 3 illustrate that the biocidal GA monomer has limited time to control microbial activity in hot and/or alkaline shales, and may return along with its aqueous transformation products to the surface via flowback water in cooler, more acidic, and saline shales.

Chapter 4 builds upon the framework set by chapter 3 to analyze another chemical commonly used in HFFs: nonylphenol ethoxylates (NPEs). NPEs are commonly used as surfactants and corrosion inhibitors in hydraulic fracturing fluids. While known to biodegrade to nonylphenol (NP), a known endocrine disrupting compound, little is known about the fate and mobility of NPEs under the extremes (temperatures, pressures, and salinities) in unconventional reservoirs. Chapter 4 presents evidence of abiotic NPE degradation directly into NP by means of hydrolysis under simulated downhole conditions (100 °C, 20 bar), revealing a previously unrecognized transformation pathway. The effects of both salinity and shale interactions were also studied, indicating that salt (NaCl) drastically accelerated hydrolysis kinetics resulting in a faster and increased production of NP, while shale induced significant sorption. Sorption to colloidal shale may result in transport of the downhole-generated NP to the surface along with the flowback and produced water. The findings presented in chapter 4 suggest that hydraulic fracturing fluids may return via flowback-produced water in a form that is more toxic than what was originally injected.

Chapter 5 of the dissertation presents the conclusions of the work presented here as well as future directions for research about downhole behavior of organic chemical additives to HFFs, using this body of work as a platform.

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CHAPTER 1 – INTRODUCTION

Rationale for Research

The technology of hydraulic fracturing (“fracking” or “fracing”) has dramatically changed the energy landscape of the world. However, there is a fog of controversy surrounding this technique, stemming from the lack of information pertaining to its potential environmental and health impacts. Though a major component of HFF is organic chemical additives, full disclosure of their identities is still not required from industry in the U.S., and thus little is known about the extent to which chemicals return to the surface in the flowback and produced water; this wastewater, which returns to the surface of a well after the well-head pressure is released, is collected upon completion of hydraulic fracturing and either reused or disposed of, making its chemical composition of particular concern, especially for risk-assessment purposes.⁶ Additionally, the fate of these chemicals in the deep subsurface (henceforth referred to as “downhole”) during a hydraulic fracturing operation has not been comprehensively studied. In fact, HFF additives are exposed to extreme chemical (e.g., salinity) and physical (e.g., temperature and pressure) conditions that may possibly result in different reactivity and transformation mechanisms as compared to near-surface environments. Consequently, flowback water may not contain the same chemicals as were originally injected, further complicating risk assessment and wastewater management.

Hydraulic Fracturing and the Downhole Environment

Fracturing a well is a fluid-intensive process, and thus large volumes of HFF additives are also being injected to maintain a target working concentration.⁶ The water used per fractured

well in the U.S. between 2010 and 2013 varied state-to-state but averaged 9,200,000 L,⁷ resulting in an average use of 46,000 L of HFF additives per well (~0.5% of the overall fluid); this translates to hundreds of millions of liters of pure chemicals injected into the subsurface every year. Currently, most research is focused on the inorganic constituents of flowback water,⁸⁻¹² which mainly originate from shale and shale-derived brine. While there are some recent studies analyzing the organic components present,¹³⁻¹⁹ many of those studies focus on organic compounds which are most likely native to the shale bitumen¹⁶ such as harmful volatile organic compounds (VOCs) like benzene, toluene, ethylbenzene, xylenes,^{15,17,19,20} and polycyclic aromatic hydrocarbons (PAHs).¹³ The few studies on organic HFF additives in flowback water have consistently identified surfactants^{14,15,21} as major organic species. In flowback and produced water samples from Colorado, Ferrer and Thurman²¹ detected the gelling agent guar gum, the biocide alkyldimethylbenzylammonium chloride (ADBAC), and aldol condensation products of the biocide glutaraldehyde (GA), but not the parent GA molecule itself.

Downhole conditions may be extremely variable between two wells, even those drilled in the same formation, exposing HFF to a wide variety of downhole conditions. For example, the Marcellus shale, which is present from 1,200 to 2,600 m in the subsurface,²² typically falls within a temperature range of 40-100 °C but can reach temperatures of 100-125 °C in the southwestern region²³ with downhole pressures typically ranging from 27,500-41,400 kPa (4,000-6,000 psi). In deeper shales such as the Haynesville in Texas/Louisiana (3,200 to 4,100 meters underground), the downhole temperature can reach almost 200 °C and corresponding pressures may be over 69,000 kPa (10,000 psi). Due to injections of acid and the buffering of carbonate species underground, the natural pH of connate water of shales and hence the pH of the mixed fluids is harder to determine; however, the pH range of flowback/produced water from

reporting states (i.e., Pennsylvania and Colorado) is between 5.1 and 8.4.^{10,15} Underground environments can also be extremely saline; it has been reported that produced water from the Marcellus contains total dissolved solids (TDS) ranging from 100,000 mg/L to 300,000 mg/L. On the other hand, some shales have comparatively lower salinities such as the Niobrara in Colorado²⁴ (Figure 4). Despite varying TDS, the predominant soluble salts present in all tested produced water samples from North American shales are Na⁺, Ca²⁺, and Cl⁻.^{8,15,24,25}

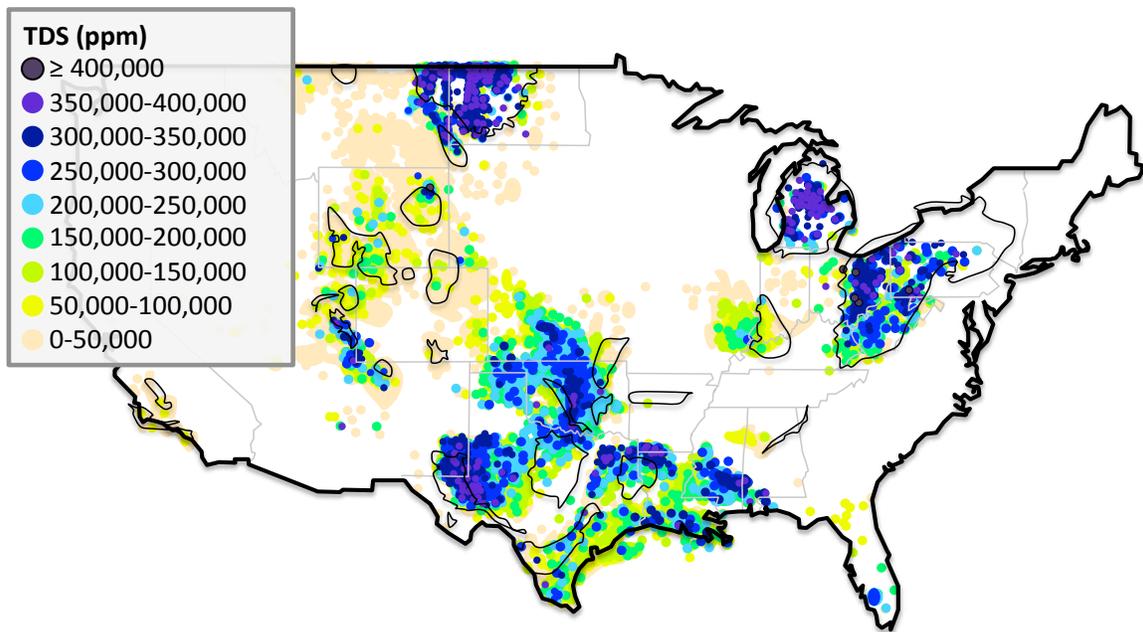


Figure 1. Produced water quality parameters such as salinity vary greatly among petroleum reservoirs. The colors above represent the total dissolved solids (TDS) in all produced water samples, respectively, gathered by the United States Geological Survey (USGS) and reported in the National Produced Waters Geochemical Database.²⁴ Traced in black over the map of the continental U.S. are outlines of all active gas and oil producing shales. Note that datasets shown include all available information on produced water samples from both conventional and unconventional petroleum recovery operations.²⁴

Chemicals Used within Hydraulic Fracturing Fluids: A Brief Overview

The chemicals used in hydraulic fracturing are added to a) alter the physical properties (such as viscosity) of the HFF to make it more effective at delivering proppant, and to b) combat

the unwanted effects of microbial activity.^{26,27} The full list of purposes that chemicals serve in HFFs includes gelling/foaming agents, friction reducers, crosslinkers, breakers (typically oxidizers that “break” the polymers), pH adjusters, biocides, corrosion inhibitors, scale inhibitors, and surfactants.^{6,27} An excellent and exhaustive review of the specific chemicals used for all of these functions was done by Elsner et. al.⁶ and hence will not be covered in depth here, while chapter two serves as the (first published) in-depth review of the biocides used in HFFs. Chemicals have historically always been used in oilfield operations– for example, in the recovery of oil, certain chemicals are needed to modify the physical characteristics of the fluid(s) being used (polymers, surfactants), while other chemicals are used to control the growth of algae and bacteria (biocides). When the industry began utilizing hydraulic fracturing to enhance hydrocarbon recovery in 1947,²⁶ they used the very same chemicals for similar purposes in the fracturing process; polymers are used to alter the viscosity of the hydraulic fracturing fluid (HFF) to optimize delivery of proppant (such as sand) into the pressure-generated cracks,²⁶ and biocides are used to kill off bacteria which may otherwise degrade the petroleum product and/or produce toxic hydrogen sulfide gas.²⁸ Many of these chemicals were traditionally meant for a closed system; that is, one with little to no chance of interaction with surface waters or other outdoor environments where the chemical could do harm (since many of the chemicals, especially biocides, are necessarily toxic). Only because of exemptions provided for hydraulic fracturing in the Safe Drinking Water Act²⁹ can many of these chemicals be used in hydraulic fracturing. Though many of these traditional chemicals have been phased out for the purpose of fracturing, many of them are still used this day.

The body of original research presented here focuses on two chemicals specifically; glutaraldehyde (GA), one of the most commonly used biocides, and nonylphenol ethoxylates

(NPEs), a chemical commonly used as a corrosion inhibitor and surfactant. Biocides, a group of reactive organic and inorganic chemicals routinely added to fracturing fluids, are used for underground microbial control to prevent the detrimental side-effects of bacterial growth and anaerobic respiration: pipe clogging due to biofilm, production of toxic H₂S gas (thus “souring” the natural gas/oil and posing a health risk to workers), and corrosion of underground casing.^{28,30-37} The problematic sulfate-reducing and acid-producing bacteria are not native to the shale, but rather are typically introduced at the surface level, either from the injected water or sand.^{28,31,32,38} Because a lethal concentration of biocide must be achieved in this injected fluid for effective microbial control, large volumes of biocides are being injected into many (but not all) of the wells being hydraulically fractured. To achieve downhole bacterial control, a variety of biocides are currently being added to hydraulic fracturing fluids (Table 1). Combinations of certain biocides may create synergistic effects, thereby decreasing the doses needed for sufficient treatment.^{39,40} Furthermore, the reactivity towards other fluid additives must be considered, as many biocides are inherently reactive molecules and side-reactions are undesirable.³³

Table 1: Chemical identification information on hydraulic fracturing biocides. Mode of action (MOA) listed is electrophilic (E), lytic (L), or oxidizing (O). Frequency of use is normalized to only those wells in which biocides were used as reported on FracFocus.¹

Trade Name & CAS No.	Chemical Structure	Chemical Formula	MOA	Freq. of Use
Glutaraldehyde 111-30-8		C ₅ H ₈ O ₂	E	27%
Dibromo-nitropropionamide 10222-01-2		C ₃ H ₂ Br ₂ N ₂ O	E	24%
Tetrakis hydroxymethyl phosphonium sulfate 55566-30-8		[(HOCH ₂) ₄ P] ₂ SO ₄	E	9%
Didecyl dimethyl ammonium chloride 7173-51-5		C ₂₂ H ₄₈ NCl	L	8%
Chlorine dioxide 10049-04-4		ClO ₂	O	8%
Tributyl tetradecyl phosphonium chloride 81741-28-8		C ₂₈ H ₅₆ PCl	L	4%
Alkyl dimethyl benzyl ammonium chloride 68424-85-1		C ₁₉ H ₃₄ NCl	L	3%
Methylisothiazolinone 2682-20-4		C ₄ H ₅ NOS	E	3%
Chloro-methylisothiazolinone 26172-55-4		C ₄ H ₄ NOCl	E	3%
Sodium Hypochlorite 7681-52-9	Na ⁺ Cl-O ⁻	NaClO	O	3%
Dazomet 533-74-4		C ₅ H ₁₀ N ₂ S ₂	E	2%
Dimethyloxazolidine 51200-87-4		C ₅ H ₁₁ NO	E	2%
Trimethyloxazolidine 75673-43-7		C ₆ H ₁₄ NO	E	2%
N-Bromosuccinimide 128-08-5		C ₄ H ₄ BrNO ₂	E	1%
Bronopol 52-51-7		C ₃ H ₆ BrNO ₄	E	<1%
Peracetic acid 79-21-0		C ₂ H ₄ O ₃	O	<1%

Not every well requires the use of biocides. Those which are particularly high in temperature, for example, may not be hospitable for microbial growth; the limit for life at surface conditions is approximately 120°C, and that is for specialized thermophiles; most surface-water

bacteria die quickly at 60°C and above. Despite this fact, bacteria remain a problem in “hot” shales ($\geq 100^\circ\text{C}$)¹⁸ and it is reported that downhole temperatures of up to 175°C (in the Horn River Basin, Canada) are insufficient to eliminate bacteria contained in source water.⁴¹ While not well documented in open literature, it is also reported that souring (production of H₂S gas) of wells in hot shales after approximately six months of production despite the presence of H₂S initially is a well-known issue in industry;¹⁸ though it is not clear exactly why this happens, microbial growth is a viable possibility. One possible explanation for the presence of harmful bacteria is the fact that the fluid injected has a significant cooling effect on the downhole temperature, and may create temperate zones downhole which may be amenable to microbial life; because of this, biocides are often used even in wells thought to be not particularly suited for bacterial growth and thus are exposed to a wide range underground environments or “downhole conditions.” This will be discussed along with biocides in further detail in chapter 2, and information specifically focused on GA (glutaraldehyde) will be revealed in chapter 3.

Surfactants, on the other hand, are used for entirely different reasons in hydraulic fracturing. Typically these chemicals are added to modify the solubility characteristics of the HFF. They are also added to modify the physical characteristics of the HFF such as to make the fluid more “slippery” along the surface of the pipeline; thus, these surfactants are also labeled as “friction reducers” such as polyethylene glycol.⁴² Still other surfactants have excellent nonpolar sorption characteristics, and since steel (which the pipeline casing is comprised of) is a nonpolar surface, these surfactants sorb to the steel surface and exist in the steel/water interface; by doing so, these surfactants create a chemical barrier which “shields” the steel surface from chemical corrosion, earning these molecules the additional label of “corrosion inhibitor.” NPE (nonylphenol ethoxylate), which will be discussed in further detail in chapter 4, is one such

surfactant. NPEs are, in fact, such powerful surfactants, they are also useful as solvents and nonemulsifiers.⁶ Their favorable physical properties have led to their widespread use; as of 2016, NPEs are disclosed in the HFF of over 50% of all hydraulic fracturing operations.⁶ Because their usefulness is independent of well depth or temperature, NPEs are exposed to nearly every downhole environment created by hydraulic fracturing. This, and the downhole behavior of NPEs, will be explored in detail in chapter 4.

Dissertation Research Objectives

The overarching objective of the following research is to examine the chemicals used in hydraulic fracturing and to study how they might transform in the unique environment provided by hydraulic fracturing operations. The fluid which returns to the surface after fracturing is complete (and pressure is released)—called “flowback” or “produced” water— is comprised of desirable hydrocarbons and also the “old” HFF and any chemicals it still contains, which is considered wastewater past this point to be either reused, recycled, or disposed of. Not knowing the underground or “downhole” behavior of the organic chemicals introduced into the HFF means that a rather critical component of the wastewater is largely not understood, hence preventing a complete risk and/or hazard assessment of hydraulic fracturing itself, and preventing proper treatment of the wastewater. Therefore this research aims to lay the framework for analyzing the organic chemicals used in HFFs and test a few model chemicals for transformation under simulated downhole conditions.

The first specific objective of the research below is to develop a system capable of mimicking the downhole environment of a hydraulic fracturing operation. The system must be capable of holding chemicals under a pressurized artificial atmosphere at high, controlled

temperatures in order to test the effects of each downhole parameter. Because salinity is a crucial factor being tested, the system must also be corrosion-resistant. The system must also enable sampling over time to enable kinetic studies. Therefore the reaction vessel must be custom-built from stainless steel parts to accommodate ports for a thermocouple, headspace control, a pressure gauge, and liquid sampling access. Specialized high-pressure valves must be installed to accommodate the high pressures needed to adequately assess this parameter. A temperature control unit must also be connected and calibrated with the thermocouple to enable temperature control via use of heat tape wrapped around the exterior of each vessel.

The second specific objective of the research presented is to test a model chemical in the reactors and analyze any chemical transformation that occurs, including a detailed kinetic analysis. GA (glutaraldehyde) was chosen due to the fact that is the most commonly used overtly toxic (being a biocide) chemical in HFFs. A detailed review of currently used biocides will provide further justification for its choice, as well as providing insight on how this biocide compares to others in use in terms of transformation potential. GA will be carefully analyzed via high performance liquid chromatography (HPLC) with ultra-violet-visible (UV-vis) detection as well mass spectrometry (MS) over time as it transforms in the reactors constructed in specific objective one, and transformation products will be identified using HPLC time-of-flight (ToF) MS. Special attention will be paid to the effect each individual downhole matrix parameter (temperature, pressure, pH, salinity, presence of shale) has on chemical transformation for the purposes of laying a testing framework.

The third specific objective of the research below is to test NPEs (nonylphenol ethoxylates) for chemical transformation in the fracturing simulators using the framework laid out by the second specific objective. The transformation of NPEs over time under simulated

downhole conditions in the reaction vessels constructed in specific objective one will be tracked using mainly HPLC-ToF-MS due to the large and aqueous nature of the analytes. Less attention will be paid to specific matrix parameters and more attention will be devoted to the transformation products themselves, which in the case of NPEs have the potential to be toxic as they could hydrolyze into the endocrine disrupting compound nonylphenol.⁶

Publication of Dissertation Work

Chapter two of the dissertation was published under the title: “Biocides in Hydraulic Fracturing Fluids: A Critical Review of Their Usage, Mobility, Degradation, and Toxicity” in the journal *Environmental Science & Technology*.²⁸ Full citation:

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Chapter three of the dissertation was published under the title “Downhole Transformation of the Hydraulic Fracturing Fluid Biocide Glutaraldehyde: Implications for Flowback and Produced Water Quality” in the journal *Environmental Science & Technology*.⁸⁸ Full citation:

Kahrilas, G. A.; Corrin, E. C.; Blotevogel, J.; Borch, T. Downhole Transformation of the Hydraulic Fracturing Fluid Biocide Glutaraldehyde: Implications for Flowback and Produced Water Quality. *Env Sci & Tech*. **2016**, *50* (20), 11414-11423. DOI 10.1021/acs.est.6b02881

Chapter four of the dissertation is currently under internal review, to be published in the journal *Environmental Science & Technology* under the name of “Endocrine disrupting chemical formation from nonylphenol ethoxylate hydrolysis under unconventional oil and gas reservoir conditions” by Kahrilas, G. A.; Blotevogel, J.; Borch, T.

CHAPTER 2 – BIOCIDES USED IN HYDRAULIC FRACTURING – A REVIEW²⁸

Introduction

Biocides are widely used in food preservation, water treatment, healthcare sanitation, textile, and other industries.⁴³⁻⁴⁶ Biocides are widely used in food preservation, water treatment, healthcare sanitation, textile, and other industries.⁴³⁻⁴⁶ During past decades, a wide variety of bioactive organic chemicals have been developed for disinfection, sterilization, and preservation purposes, including quaternary ammonium compounds, alcoholic and phenolic compounds, aldehydes, halogen-containing compounds, quinoline and isoquinoline derivatives, heterocyclic compounds, and peroxygens.^{47,48} Biocides have also been applied in oil reservoirs for many decades, particularly in water flooding operations during secondary oil recovery.³⁶ Likewise, biocides are among the most common chemical additives used for hydraulic fracturing (“fracking”, or “fracking”), a process in which a water-based fluid is used to help induce cracks in oil- and/or natural gas-containing unconventional formations such as shale rock. At total concentrations of up to >500 mg/L⁴⁹ and total fluid volumes surpassing ten million liters per horizontal well,⁵⁰ total amounts of biocide(s) used per hydraulic fracturing event can exceed 1,000 gallons.⁵¹

Bacterial control is necessary in hydraulic fracturing operations to prevent excessive biofilm formation downhole that may lead to clogging, consequently inhibiting gas extraction.³⁰ Biocides inhibit growth of sulfate-reducing bacteria (SRB),^{31,32,38} which anaerobically generate sulfide during the organisms’ respiration process. Sulfide species created in the subsurface may pose a risk regarding occupational safety and health when the fluid returns along with produced

H₂S gas. Furthermore, SRB and acid-producing bacteria (APB) may induce corrosion of the production casing / tubing underground, potentially leading to casing failure and environmental contamination by petroleum products.³¹⁻³⁷

Hydraulic fracturing operations provide bacterial species with many habitats favorable to their (unwanted) growth and proliferation. The major sources of bacterial contamination are (1) drilling mud, (2) water, (3) proppants, and (4) storage tanks. Prolonged storage of water prior to use, typically in lined or unlined earthen pits,^{31,32} can lead to mass proliferation of microorganisms. Likewise, bacteria can thrive in stored produced water that was recycled for use in future fracturing operations.^{23,52} The increased temperatures fracturing fluids are exposed to underground may also favor microbial growth,⁵³ and therefore many bacterial species (including anaerobic species that are native to shale formations)^{34,54-57} may proliferate underground during hydraulic fracturing. A diverse array of bacteria including those within the taxa *γ-proteobacteria*, *α-proteobacteria*, *δ-proteobacteria*, *Clostridia*, *Synergistetes*, *Thermotogae*, *Spirochetes*, *Bacteroidetes*, and *Archaea* have all been found in untreated flowback water samples.⁵⁷ Sulfate reduction by piezophilic bacteria coupled to oxidation of methane or organic matter is stimulated at high pressure;⁵⁸⁻⁶² pressure increases naturally underground but is also artificially increased during the process of hydraulic fracturing, matching values relevant to the referenced studies. Similar pressure-stimulation effects have also been observed in iron-reducing bacteria.^{58,63,64} In fact, reduction in viability of bacteria due to high pressure does not occur until 100-150 MPa (~15,000-22,000 psi),⁵⁸ which exceed most formation pressures.

Biocides are often (but not always) used in hydraulic fracturing fluid formulations. While their application is often mandatory in aboveground oil–water separation units, water storage tanks, and pipelines used to transport these fluids, it is believed that the extremely high

temperatures inherent to some shale formations may naturally impede microbial growth.³⁶ The temperature of shale plays in the continental U.S. varies greatly, even within a single formation. The gas-bearing shales of the Marcellus Formation, which exist from 1,200 to 2,600 meters underground,^{22,65} typically fall within a temperature range of 40-100 °C, but can reach temperatures of 100-125 °C in the southwestern region.²³ In deeper shales such as the Haynesville in Texas/Louisiana (3,200 to 4,100 meters underground⁶⁵), the downhole temperature can reach almost 200 °C.^{65,66} However, a few studies have suggested that some bacteria are very persistent and may not be completely killed by the extreme underground conditions,^{41,55,66} or that higher pressures may prevent bacterial death at higher temperatures.^{61,67} Furthermore, injection of colder fracturing fluids may lead to considerable cooling of the casing and target formation. Thus, biocides are sometimes added to fracturing fluids even in formations with temperatures exceeding 122 °C, the highest recorded temperature at which aerobic bacteria reproduction has been observed.⁶⁸

Unlike water flooding, in which there are continuous inflows and therefore a need for long-term suppression of microbial activity, the goal of biocide application in hydraulic fracturing is to reduce the deleterious microbial populations up-front to the lowest levels possible, in order to mitigate the risk of their colonization of the reservoir and well system long after the fracturing operation has been completed. This is due to the fact that reservoir souring in the fractures downhole cannot be cured after hydraulic fracturing is complete.⁶⁹ While sterilization is unlikely, targeting deleterious bacteria with biocides that have been efficacy-tested for performance against those specific species can lead to long term protection for months or longer, mitigating the risk for souring and microbially induced corrosion (MIC). Thus, it is critical to understand microbial dynamics in this context, i.e., types and concentrations of

microorganisms, carbon sources, nitrogen sources, and electron acceptors present, as well as growth limiting factors. Furthermore, potential growth rates of microorganisms in fracturing fluids under subsurface conditions need to be considered, which are not yet fully understood.⁵⁵⁻⁵⁷ To determine a suitable site-specific biocide (combination), 6-log reductions or greater in SRB, APB, or other deleterious bacterial populations are typically aimed for in preliminary laboratory testing according to NACE Standard TM0194.⁷⁰ When choosing suitable biocide(s), typical specific parameters of the fracturing operation that can affect biocide performance are taken into account. Some of these parameters include compatibility with the other fracturing chemicals being used, flow rates of the fracture fluids, and the chemistry of the water used. Reservoir conditions such as temperature and formation geology may also be considered. However, selection of biocides is often done based on historical precedence, and currently is not optimized on a well-by-well basis.

The following sections of this review will focus on the most common hydraulic fracturing biocides by frequency of application (Table 1) according to the national hydraulic fracturing chemical registry FracFocus.¹ FracFocus is managed by the Ground Water Protection Council and the Interstate Oil and Gas Compact Commission, and is currently used as the official state chemical disclosure system in ten U.S. states (CO, OK, LA, TX, ND, MT, MS, UT, OH, PA). While the registry is not inclusive of wells outside the listed states and is dependent on company-volunteered information in states that do not mandate reporting of injected chemicals, it is the largest registry of its type and allows an approximate estimation of national chemical use trends. Additionally, not all compounds listed as biocides on FracFocus are active ingredients with biocidal activity (i.e., ethylene glycol); regardless, these compounds are registered as biocides because they are components in commercial biocide product mixtures. To enable

assessment of the potential environmental and health impacts of the biocides used in hydraulic fracturing fluids, we review their relevant pathways of environmental contamination, environmental mobility, stability and chemical behavior in a variety of natural environments, and toxicity. Alternatives to traditional chemical biocides will be highlighted as methods of achieving the necessary bacterial control in hydraulic fracturing operations while lowering environmental risks. Finally, areas in need of research are revealed as current knowledge gaps impede full understanding of environmental fate and transport of biocides.

Hydraulic Fracturing Biocides and their Modes of Action

Biocides function by various modes of action to control bacteria – they are generally divided into oxidizing and non-oxidizing compounds.⁷¹ Oxidizing biocides such as bromine- (e.g., N-bromosuccinimide, NBS) and chlorine-based (e.g., chlorine dioxide and sodium hypochlorite) species rely on the action of released free radical species that attack cellular components.^{72,73} Peroxides are often used in pre-treatment of obtained natural water sources due to the fact that they are extremely non-specific and work efficiently on a wide range of bacteria.^{73,74} However, oxidizing biocides pose the risk of equipment corrosion as well as unwanted reactions with other hydraulic fracturing chemicals. Furthermore, their reaction may produce halogenated hydrocarbons and other unwanted disinfection byproducts (DBPs).^{33,75} Oxidizing biocides are too short-lived to control microbial growth for long periods of time during fluid storage, and to control growth of bacteria that may already be present in the well bore area from the drilling operation, making this class of biocide poorly suited for injection.⁷⁶ For these reasons, oxidizing biocides are more commonly used for on-site treatment of stored fresh and wastewater, and less often in hydraulic fracturing fluids (Table 1, SI Figure S2)

(though their use is increasing in some areas based on trial-and-error). However, oxidizing biocides are well-reviewed in existing literature, and therefore will not be emphasized in this critical review.

The most common biocides used in hydraulic fracturing fluids used are non-oxidizing organic chemicals, which will therefore be focused on. These biocides are split into two groups according to their respective modes of action: electrophilic, and lytic (often used in conjuncture in fracturing fluids). Many of these biocides are not exclusive to hydraulic fracturing and are used by many other industries and products.

Lytic Biocides. Lytic (also known as membrane-active) biocides are amphiphilic surfactants and their activity is generally based on dissolution into the bacterial cell wall and its subsequent disruption.⁷⁴ Specifically, their known mode of action involves binding to anionic functional groups on the membrane surface and subsequent perturbation and dissolution of the lipid bilayer, resulting in loss of osmotic regulation capacity and eventual lysis of the cells.⁷⁷ The two main lytic biocides used for hydraulic fracturing are the cationic quaternary ammonium/amine compounds ("QACs", or "quats") didecyl dimethyl ammonium chloride (DDAC, or decanaminium, Table 1) and alkyl dimethyl benzyl ammonium chloride (ADBAC, benzalkonium chloride, BAC, BC, or benzenemethanaminium), although they are often not distinguished specifically on disclosure forms. These biocides are characterized by a central quaternary nitrogen atom that carries a permanent positive charge and is bonded to four carbon-containing "R"-substituents.

QACs are injected into 22% of all wells registered on FracFocus and used in nearly every shale formation in the U.S.,¹ though not solely as biocides since QACs also serve as cationic surfactants, corrosion inhibitors, and clay stabilizers.⁷⁸ The QACs used for biocidal purposes,

DDAC (comprising 8% of all biocide use, Table 1) and ADBAC (3% of all use, Table 1), are often combined with electrophilic biocides such as glutaraldehyde to increase their efficiency through synergistic effects thus lowering the total amounts of biocide needed for appropriate bacterial control.^{1,79} The only other commonly used lytic biocide is tributyl tetradecyl phosphonium chloride (TTPC), whose U.S. EPA registration review is scheduled to be completed in 2017.⁸⁰ Therefore, information on this substance is limited.

Electrophilic biocides. Electrophilic biocides typically have reactive electron-accepting functional groups (i.e., aldehydes) that react with electron-rich chemical groups such as exposed -SH (thiol) and -NH (secondary amine) groups in membrane proteins on bacterial cell walls.⁷⁴ Glutaraldehyde ("glut") is the most commonly used electrophilic biocide in hydraulic fracturing operations (Table 1). Similar to formaldehyde and other aldehydes, it is a potent cross-linker for amino and nucleic acids (SI Scheme S1), leading to cell wall damage and cytoplasmic coagulation.⁸¹ Glutaraldehyde is one of the main biocides being used in off-shore hydraulic fracturing operations internationally.⁸²

Both 2,2-dibromo-2-nitropropionamide (DBNPA, 2,2-dibromo-2-cyanoacetamide) and 2-bromo-2-nitropropane-1,3-diol (Bronopol) are not considered oxidizing biocides despite small releases of bromine⁸³ which are assumed to enhance their biocidal activity.^{71,84} Both rapidly react with sulfur-containing nucleophiles such as glutathione or cysteine, thereby disrupting key cell components and biological functions.⁸⁵

Another quaternary phosphonium compound currently registered for use as a biocide in hydraulic fracturing fluid is tetrakis hydroxymethyl phosphonium sulfate (THPS). Under alkaline conditions, it deformylates and releases trihydroxymethyl phosphine (THP), which cleaves sulfur-sulfur bonds in the disulfide amino acids of the microbial cell wall⁸⁵ Dazomet (3,5-

dimethyl-1,3,5-thiadiazinane-2-thione, or “mylon”) is often combined with THPS in fracturing fluids.³²

Finally, although rare, use of sulfur-containing biocides is still reported (i.e., California and Texas, Table 1 and Figure 3). The most commonly used sulfur-containing biocide treatment is comprised of two chemicals, chloromethylisothiazolinone (CMIT, or MCI, 5-chloro-2-methyl-3(2H)-isothiazolinone) and methylisothiazolinone (MIT, or MI, 2-methyl-3(2H)-isothiazolinone). They react with a wide range of amino acids, inhibiting critical metabolic processes.

Modes of Accidental Environmental Contamination

Environmental exposure to biocides after an inadvertent release may occur during 1) transportation of chemicals to well pads (Figure 1, pathway 1); 2) mixing of chemical additives with the bulk of the fracturing fluid (pathway 2); 3) injection of the mixed fluid into the borehole (often occurs simultaneously with in-line mixing) (pathway 2); 4) handling, collection, and storage of chemical-containing produced water (pathways 5 and 6); and 5) reuse, treatment, recycling, and/or disposal of the produced water (pathways 3, 4, 5, and 7).⁸⁶ The hydraulic fracturing event itself may have a considerable impact on the chemical stability of the organic additives and thus on the composition of the produced water. As the fluids are exposed to the high pressures, temperatures, salt concentrations, and organic matter contents of the oil- and gas-bearing formations, abiotic transformation and sorption reactions may greatly accelerate the chemicals' depletion (Figure 2). To our knowledge, there is currently only one study that directly addresses this critical effect.⁸⁷ For the biocide glutaraldehyde, it was shown that transformation increases with increasing temperature and pH, and may be substantial under conditions encountered downhole.⁸⁸ Consequently, it can be expected that a portion of the injected biocides

will not resurface, or may resurface as transformation product(s). Thus, the potential risks associated with biocide exposure may differ substantially before and after hydraulic fracturing.

Few studies have been conducted on the presence of organics in produced water from shales which had undergone hydraulic fracturing.^{13,14,89} The compounds detected included polyaromatic hydrocarbons (PAHs), aliphatic hydrocarbons, and long-chain fatty acids.¹³ Certain chemicals unique to hydraulic fracturing, including ethoxylated surfactants¹⁴ and the biocide 1,3,5-trimethylhexahydro-1,3,5-triazin-2-thione (a Dazomet derivative),¹³ were also detected in produced water. Levels of dissolved organics in the produced water as measured directly from active wellheads decreased sharply after the first 20 days; concentration of the biocide dropped from 1.5 mg/L on day 1 to 0.01 mg/L on day 8.¹³

The most commonly reported accidents leading to environmental contamination are surface spills (pathways 1-3 in Figure 1), which can happen on-site (including well blowouts and casing failures)^{51,90,91} or during transportation to or off the site via pipelines, trains, or trucks.⁹²⁻⁹⁴ On-site spills, which may result in surface water, soil, or shallow groundwater contamination (pathways 5-6 in Figure 1),⁷⁵ are often a result of use of lined pits to temporarily store and evaporate flowback brine in order to reduce the volume of waste.^{51,90} More recently, storage tanks have replaced these pits in an attempt to reduce the potential for unintentional spills.⁹⁵

While comprehensive and complete data on surface spills for most shale plays are still lacking or are otherwise difficult to access, the Colorado Oil and Gas Conservation Commission (COGCC) offers a robust database that includes any spill or release of exploration and production waste within the State of Colorado of one barrel or more outside, and of five barrel or more inside of berms or other secondary containment.⁹⁶ In 2013, there were 591 reported spills, which released a total of 14,067 barrels (i.e., ~2,200,000 liters), or 0.004% of all produced water.

This relates to a total of 50,067 active oil and gas wells in Colorado, including 4,025 new wells that had been drilled in 2013.

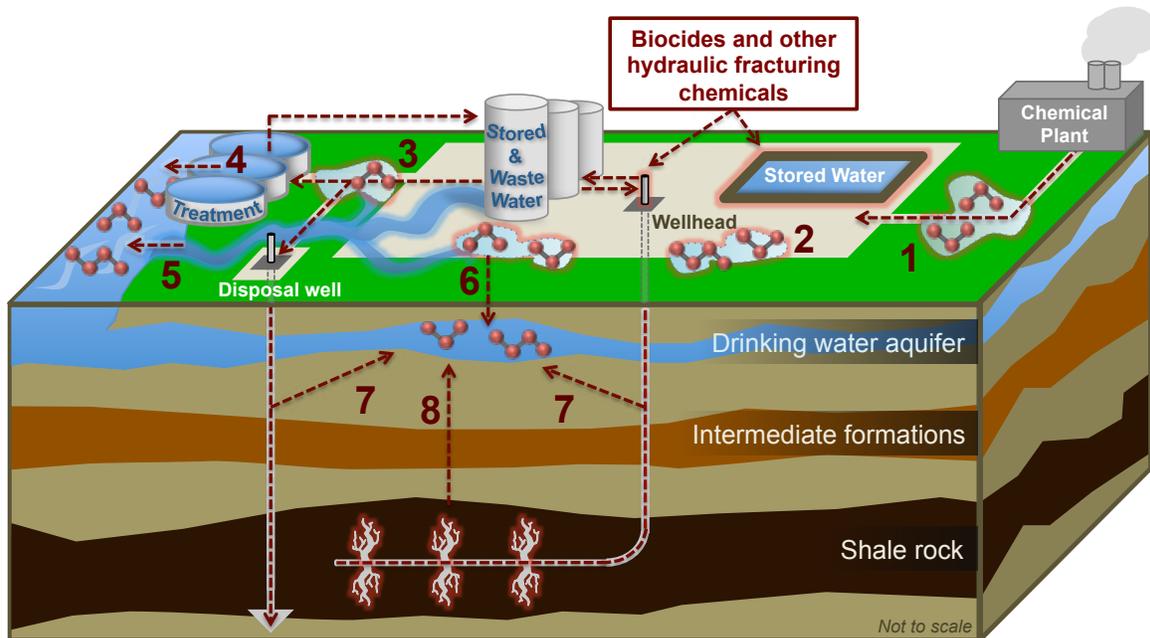


Figure 2: Modes of potential environmental exposure: 1-3) surface spills resulting in contamination of soil; 4) incomplete removal in treatment plants; 5) surface spill runoff into surface water; 6) surface spills leaching into shallow aquifer; 7) contamination of shallow groundwater via borehole leakage, fault lines, and abandoned wells; 8) contamination of shallow groundwater via induced fractures. Artwork is conceptual and not drawn to scale.

To minimize wastewater volumes and thus potential environmental impacts, wastewater can be reused in subsequent hydraulic fracturing events, which is the dominant management practice in the Marcellus Shale region.⁹⁷ Of the 12,604 instances of unconventional waste disposal (which includes all fluid and solid waste generated from unconventional resource extraction) reported in Pennsylvania from January to June in 2013, 33.4% indicated treatment by a centralized plant and then recycled in another hydraulic fracturing operation, 30.3% indicated direct reuse of the wastewater in other drilling operations, 26.0% indicated disposal via injection in disposal wells, 9.5% indicated disposal via landfills, and only 0.3% indicated treatment for eventual discharge

into surface waters.⁹⁸ It is important to note that wastewater treatment strategies vary between states and these statistics may not necessarily be representative of those nationwide.

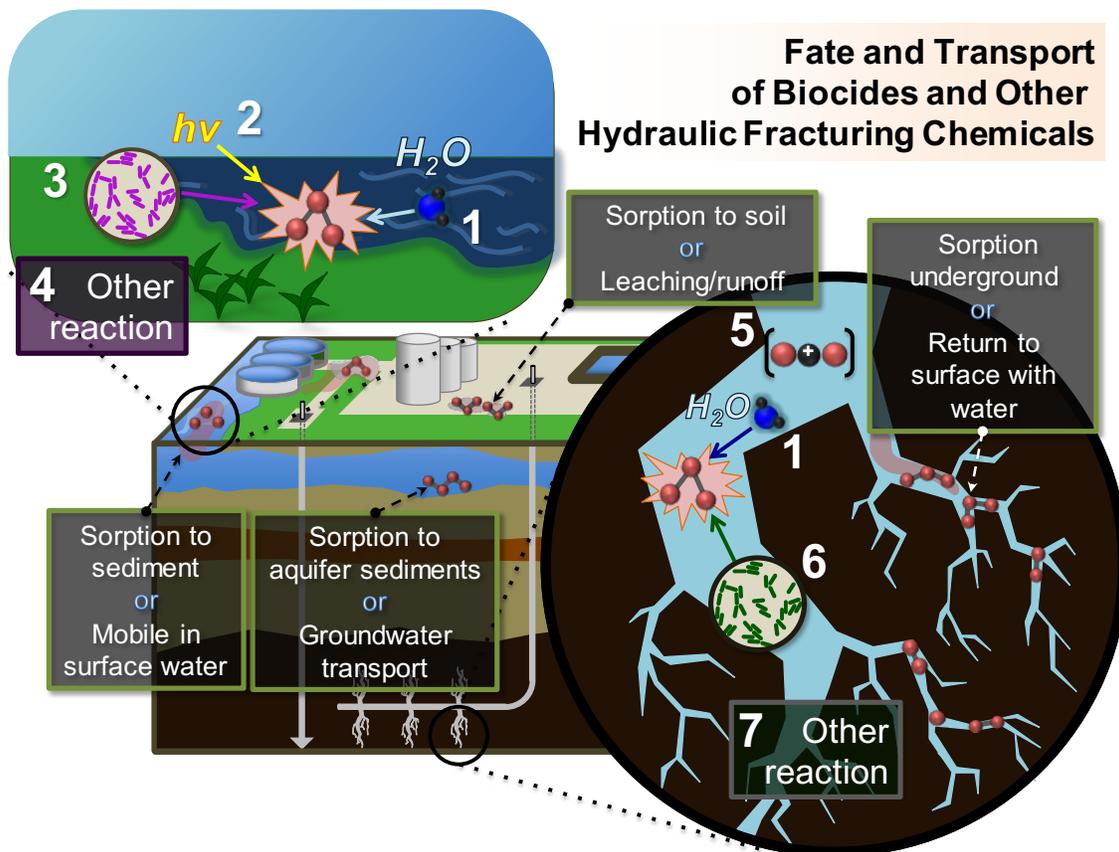


Figure 3: Fate and transport of hydraulic fracturing chemicals (including biocides) in different environments. Possible degradation pathways are numbered as follows: 1) hydrolysis; 2) direct or indirect photolysis; 3) aerobic biodegradation in the water or soil; 4) other chemical reaction with oxygen present; 5) complexation underground with dissolved inorganic species; 6) anaerobic biodegradation; 7) other chemical reaction (e.g., nucleophilic substitution or polymerization) under anoxic conditions, high pressure, and elevated temperature. Artwork is conceptual and not drawn to scale.

While injection into deep underground reservoirs is currently the most common method of unconventional waste disposal,^{99–101} little is known about the long-term impact and risk of this technique.^{99,102} However, depending on exposure times of the organic chemical additives to the increased pressures and temperatures of these deep formations, (further) chemical transformation is conceivable.⁸⁷ Similar to fracturing operations, environmental exposure to the injected solution

may occur if the cement or casing is faulty (pathway 7, Figure 1); however, this has yet to be documented.

In the minority of instances where wastewater is treated for discharge, publicly owned treatment works (POTWs), municipal wastewater treatment plants (WWTPs), or commercially operated industrial wastewater treatment plants are used to treat hydraulic fracturing wastewater.^{98,103} However, most of the municipal treatment plants are designed for common compounds (nutrients and organic matter), and were not intended to treat the multitude and amounts of chemical species that resurface with flowback brine. Furthermore, assuming the presence of biocides in wastewater at sublethal concentrations, adaptation of surviving microorganisms to biocides becomes possible – a process that is well documented for ADBAC^{104,105} and other biocides.^{40,106} This biocidal resistance is sometimes accompanied by low-level increases in antibiotic resistance,¹⁰⁷ which is already an existing problem in WWTPs.¹⁰⁸ Fortunately, dilution below the minimum inhibitory concentration also enables the majority of these chemicals to undergo biodegradation in WWTPs and in natural waters, which will be discussed in more detail below. In hypothetical cases where biocides persist through WWTP treatment, however, contamination of water and soil may occur (pathway 4 in Figure 1).

Although WWTP effluent is typically released into natural streams and waterways, there are rare documented cases of its reuse for other purposes such as crop irrigation,^{94,109} or spread on roads to reduce dust and/or deice.^{75,98,110,111} Finally, wastewater from hydraulic fracturing operations off the coast of southern California is sometimes discharged directly into the ocean; as of recently, the EPA is requiring full disclosure of all chemicals meant for direct discharge into the Pacific Ocean.¹¹²

In contrast to accidents occurring at the surface, underground contamination phenomena may go unreported and undetected for years before any negative consequences are observed. As such, though not considered a dominant exposure pathway in hydraulic fracturing operations, underground exposure pathways are the focus of much peer-reviewed literature. The majority of research available suggests that natural upward migration of chemicals such as biocides from hydraulically induced fractures to aquifers (Figure 1, pathway 8) is unlikely.^{113,114} Usually, hydraulic fracturing is performed >1,000 meters below a shallow aquifer used for drinking or irrigation water extraction. Even though some fractures have been documented to grow upward by as much as 450 meters, their tops were still almost 1,500 meters below the aquifer.¹¹⁵

Underground contamination via borehole leakage, preexisting vertical cracks, fault lines, or uncharted boreholes (Figure 1, pathway 7) remains a possibility. Several studies using isotopic analysis have traced methane found in groundwater back to natural gas wells nearby.¹¹⁶⁻¹¹⁹ Darrah and co-workers (2014)¹¹⁴ demonstrated that fugitive gas contamination was due to (1) release of intermediate-depth gas along the well annulus, likely due to cement failure, (2) release of target formation gas implicating improper, faulty, or failing production casings, and (3) underground well failure. While upward flow of leaking hydraulic fracturing fluids would be substantially slower than that of buoyant natural gas⁷⁵, production well failure in the proximity or above an aquifer is a more likely potential pathway for groundwater contamination by fracturing fluid components. However, due to the lack of solid (baseline) data, further research is needed to address this question.

In conclusion, while a realistic potential for groundwater contamination via natural upward migration of deep-injected biocides may exist if the fractured formation is very shallow, it appears unlikely.^{97,114} However, several other scenarios exist which may result in unintentional

environmental contamination with biocides, including surface spills and well integrity issues associated with casing or cement failure.^{75,86,97,114,119–121}

Mobility

Table 2 summarizes the key physicochemical properties that can be used to predict the mobility of the hydraulic fracturing biocides. The property estimation software EPI Suite v4.11² was chosen after statistical comparison of the performance of different estimation methods (SI Table S3) to close the large existing experimental data gaps regarding their water solubilities, Henry's law constants (K_H), soil organic carbon-water partition constants (K_{OC}), and octanol-water partition constants (K_{OW}). Meaningful pKa values were not found in the literature.

In general, it can be seen that the organic-based hydraulic fracturing biocides considered here possess low K_H values, and are thus not very volatile. Consequently, partitioning into the air phase is not considered a major pathway. However, biocides tend to be water-soluble or miscible, enabling their transport via surface water or groundwater depending on their affinity for sorption to natural solid phases (Table 2, Figure 2).

With the exception of glutaraldehyde ($\log K_{OC} = 2.07 - 2.70$),¹²² the K_{OC} values of the uncharged biocides lie predominantly below 100 (i.e., $\log K_{OC} < 2$), indicating comparably low retardation during aqueous transport through the soil, especially soils which are low in organic matter content. Glutaraldehyde's mobility is reported to be moderate in soils and high in sediments,¹²² and due to its rapid aerobic degradation in soil and water (see below), its potential for soil contamination may only be relevant under anaerobic conditions.

.Table 2: Physical constants of biocides used in hydraulic fracturing operations. Values in bold italics are calculated using EPI Suite v4.11.² Biodegradation reported in aqueous phase (aq) or soil under aerobic (aer) or anaerobic (anaer) conditions. *Estimation uses MCI method (otherwise K_{OW} is used in K_{OC} predictions).

Biocide	log K_{ow}	Water sol. @25°C (mg/L)	Henry's coefficient, log K_H	log K_{oc}	Hydrolysis half-life (25°C)			Photolysis half-life (12 h day)	Readily biodegradable (half-life < 30 days)
					pH 5	pH 7	pH 9		
ADBAC	no data	>350,000 ¹⁰⁰	-10.87	5.810 ¹⁰⁰	stable ¹³⁰			stable (aq) ¹³⁰	no ¹³⁰
Bronopol	-0.64	200,000 ²¹⁹	-10.88 ²¹⁸	-0.033	1.5 y (pH 6 20°C) to 2 months (pH 8, 20°C) ¹³⁴			2 d (aq, pH 4) ¹³⁴	no data
Chlorine dioxide	-3.22 ²²¹	3.01E+03 ²²⁰	-1.40 ²²⁰	no data	<25 min ²²¹			no data	no data
CMIT	-0.34	149,000	-7.45	0.711	stable		22 d ²²³	no data	aq(aer) ²²³
Dazomet	0.63 ²¹⁸	3,000 ²¹⁸	-9.30 ²¹⁸	1.000 ²¹⁸	23.8 h	180 h	45.6 h ¹⁴⁴	1.6 h (aq,pH7) ¹⁴⁴	no data
DBNPA	0.82 ²¹⁸	15,000 ²¹⁸	-7.72 ²¹⁸	1.767	67 d	63 h	73 min ¹⁶⁸	14.8 h (aq,pH5); ¹⁶⁹ 6.9 h (aq,pH7)	aq(aer,anaer) ¹⁶⁹
DDAC	≈0 ¹³¹	700 ²²⁶	-9.16	5.64-6.20 ¹⁰¹	368 d	184.5 d	506 d ¹²⁹	132 d (soil) ¹²⁹	no ¹²⁹
DMO	0.73 ²²⁸	miscible ²²⁷	-5.52	0.747	<5 min ²²⁸			no data	no data
Glutaraldehyde	-0.33 ⁹⁹	miscible ²²⁹	-4.48 ⁹⁹	2.07-2.70 ⁹⁹	628 d	394 d	63.8 d ⁹⁹	196 d (aq pH 5) ⁹⁹	aq(aer,anaer) ⁹⁹
Hypochlorite	-3.42 ²²¹	miscible ²³⁰	non-volatile ²²¹	no data	<25 min ²²¹			no data	no data
MIT	-0.83	536,700	-7.30	0.440	stable		22 d ²²³	no data	aq(aer) ²²³
N-Bromo	-1.19	14,700 ¹⁸⁸	-6.41	0.259	unstable, no data			no data	no data
Peracetic acid	-1.07	miscible ²³³	-5.67 ²¹⁸	0.104		8.295 h	²³⁴	8.3 days ²³⁵	aq(aer,anaer) ²³⁵
THPS	-9.77	400,000 ²¹⁸	-12.64	-5.302	131 d	72 d	7 d ¹³⁹	<2 h (aq,pH7) ¹⁶⁶	aq(aer,anaer), s(aer) ¹⁶⁶
TTPC	no data	miscible ¹⁹⁴	3.75*10 ⁻⁸ mm Hg, 39°C ¹⁴⁵	7.66*	unknown			no data	no data
TMO	(≈DMO)	(≈DMO)	-5.18	0.841	<5 min ²²⁸			no data	no data

In contrast, ADBAC, DDAC, and TTPC possess substantially higher log K_{OC} values between 5.6 and 7.7,^{2,123,124} which is due in part to their large hydrophobic moieties.¹²⁵ In addition to hydrophobic interactions with soil organic matter, these cationic biocides can adsorb to permanent negatively charged clay surfaces, an effect which enables the use of these compounds as shale inhibitors/clay stabilizers (only quaternary ammonium/amine polyelectrolytes called “polyquats” with multiple charges are used for this purpose).⁷⁸ Despite being sorbed, these biocides potentially remain bioactive.¹²⁶ Several studies reported strong sorption of cationic organic amines to the surfaces and interlayers of clay beyond its cation exchange capacity, causing extensive clay aggregation.^{127–129}

Experiments simulating river-water contamination revealed that the quaternary ammonium compound paraquat (*N,N'*-dimethyl-4,4'-bipyridinium dichloride) becomes almost

completely sorbed onto sediments ($\geq 97\%$).¹³⁰ Similar findings were reported from leaching studies with CTAB (cetyltrimethylammonium bromide), which showed strong sorption to silica and soil.^{131–134} Moreover, it was observed that adsorption of CTAB to silica surfaces occurs within minutes, indicating that QACs will quickly sorb to soils and sediments upon accidental release.¹³¹ This may result in the accumulation of chemical in soil or sediments affected by a spill.⁷⁵

Studies investigating QACs in municipal sludge observed a substantial extent of sorption.^{135,136} Since about 50% of the municipal biosolids in the US are land-applied,¹³⁵ an accumulation of QACs in agricultural soils over time may result, potentially leading to plant uptake as is seen with other pollutants such as organophosphates,¹³⁷ or to contamination of water resources through leaching and runoff.

Sorption of hydraulic fracturing biocides (and other organic additives) may also occur downhole in the resource-bearing fractured formations. Though unconventional formations such as shale are typically non-permeable, the process of hydraulic fracturing induces many micro-fractures, which drastically increases the number of available sorption sites. Negatively charged sorption sites, to which cationic species such as DDAC and ADBAC are strongly attracted to, are abundant in shale rock due to the presence of clays.¹³⁸ Furthermore, the total organic carbon content in unconventional shale formations can exceed 10%.¹³⁹ Thus, partitioning into organic carbon-dominated phases (e.g., kerogen) even for biocides with a rather low $\log K_{oc} < 2$ may become a major pathway for their removal under downhole conditions.¹²⁵ It has also been shown that elevated temperature increases the rate of chemical sorption of organics to soils, sediments, and clays¹³⁸ within time-scales relevant for hydraulic fracturing operations. When further

considering the high pressures (known to enhance adsorption processes^{125,140}) under downhole conditions, substantial sorption of hydrophobic biocides underground can be expected.

The presence of high salt concentrations may have a considerable impact on biocide sorption to organic and inorganic surfaces. The presence of salt is partly a result of injection of acid into the well prior to injection of the organic fracturing additives. This acid, usually hydrochloric (HCl), is used in concentrations reaching 20% in the injected fluid¹ and induces dissolution from the shale, including iron, calcium, and radium.^{8-10,141} For the uncharged biocides, increasing sorption with increasing salt concentration can be expected due to decreasing solubility as a result of salting out.¹⁴² For cationic biocides, which may sorb via electrostatic interactions in addition to hydrophobic forces, the effect of salt concentration is more complex as the salts increasingly compete for ion exchange sites, thus counteracting the salting-out effect. Several studies observed that the sorption affinities of QACs to both clays and organic matter generally decreased with increasing salt concentration.^{129,143,144}

The potential for bioaccumulation of the neutral organic biocides is predominantly low as their K_{OW} values consistently lie below 10 (i.e., $\log K_{OW} < 1$, Table 2), for many even below 1 (i.e., $\log K_{OW} < 0$), indicating a preference for the aqueous phase over the organic. However, as cationic surfactants typically accumulate at the interface between two phases and may be attracted to the negatively charged surface of cell membranes, the K_{OW} is not sufficient to predict bioaccumulation of ADBAC, DDAC, and TTCP. On the other hand, their potential to undergo strong ionic bonding may limit their mobility, and thus their potential for uptake by organisms: QACs have been found to be far less bioavailable (though potentially still bioactive) to organisms than other pollutants having similar K_{OW} values,^{145,146} such as bisphenol-A¹⁴⁷ and organophosphates.¹³⁷

Degradation

Lytic biocides have been reported to be relatively resistant to degradation in a wide range of environments. They are stable in the presence of strong oxidants,¹⁴⁸ under acidic and basic conditions,¹⁴⁸ and when exposed to heat,⁷⁶ and are not readily degraded by hydrolysis, photolysis, or bacteria.^{149–151} On the other hand, electrophilic biocides are reactive chemicals, and are therefore relatively short-lived in natural environments where reduced functional groups of proteins and organic matter are prevalent. None of the chemicals react with themselves with the exception of glutaraldehyde (SI Scheme S3): unique among biocides, glutaraldehyde undergoes auto-polymerization via aldol condensation, especially at high pH values.¹⁵² The resulting α,β -unsaturated polymer is not considered toxic, nor are the products of most known reactions between these biocides. Bronopol is an exception as it reacts with oxygen and thiols to produce superoxide, a reactive oxygen species.⁸³

Hydrolysis. Hydrolysis is a major degradation pathway common to electrophilic biocides. Characterized by the addition of a water molecule resulting in two smaller fragment molecules, this process is strongly affected by the pH of the surrounding environment. In some cases, the hydrolysis products can be more toxic and/or persistent than their parent compounds, which is the case with the biocides DBNPA,^{84,153} Bronopol,^{154–158} THPS,^{32,159–161} and Dazomet.^{32,162–164} DBNPA's major products via hydrolysis (which occurs readily above pH 8.5⁷⁴) are dibromoacetic acid and dibromoacetonitrile (DBA, SI Scheme S4).^{84,153}

Bronopol hydrolyzes within 3 hours at 60 °C and pH 8, producing formaldehyde, nitrosamines, and other molecules.^{154–158} Although the parent compound (Bronopol) is rather short-lived in the environment, its degradation products are toxic and more persistent.^{155,165} The hydrolysis degradation products of THPS are trihydroxymethyl phosphine (THP) molecules that

subsequently degrade into trishydroxymethyl phosphine oxide (THPO), accompanied by the release of two formaldehyde molecules.^{32,159–161} Dazomet's hydrolysis half-life in water of pH 7 is 7.5 days (Table 2).¹⁶² Its major products are methyl isothiocyanate, formaldehyde, monomethylamine, hydrogen sulfide, carbon disulfide (in acid soils), and nitrogen oxides.^{32,162–164}

Another influence on chemical stability underground is the pH of the surrounding environment. Analysis of produced water revealed a pH range of 5-7, controlled mainly by bicarbonate species intrinsic to the shale formations.⁹ Although the biocides used in hydraulic fracturing are relatively stable at near-neutral pH, small changes in pH can nonetheless have a large impact on any acid- or base-catalyzed reactions.^{87,88}

Photodegradation. Some of the hydraulic fracturing biocides contain chromophores, such as MIT, CMIT, Dazomet and N-Bromo, and may thus undergo direct photolysis reactions when exposed to ultraviolet (UV) light. Experimentally, this has thus far only been tested for DBNPA and Bronopol. Via direct photolysis, DBNPA yields dibromoacetic acid as the major degradation product (SI Scheme S5),⁸⁴ Bronopol was shown to produce tris(2-hydroxymethyl-2-nitropropane-1,3-diol).¹⁵⁴ In addition, all biocides can be degraded by the action of reactive species from indirect photodegradation. However, no experimental data are available for this process.

Biodegradation. Microbial degradation and mineralization of biocides becomes possible upon dilution or degradation to sub-lethal levels, and/or due to proliferation of biocide-adapted bacteria.^{54,55,104} Minimum inhibitory concentrations (MICs) not only depend on the type and mixture of biocide(s) but also on the type of organism, and are typically lower for planktonic

cells compared to biofilms.³⁴ For many hydraulic fracturing biocides, typical MICs are on the order of milligrams to hundreds of milligrams per liter.³⁴

Below MICs, bacterially-induced degradation of biocides can occur in soil or surface water in the event of a spill, underground as a result of ineffective bacterial control, or at WWTPs.^{166,167} In soils, it is generally concluded that cationic surfactants such as DDAC, ADBAC, and TTPC exhibit such strong sorption to soil that their bioavailability and thus biodegradation rates are substantially reduced.^{149,150,168,169} One strain of *Pseudomonas fluorescens* TN4, which had been isolated from activated sludge from a WWTP, was shown to be not only highly resistant to DDAC, ADBAC and other QACs, but also capable of aerobically degrading them at concentrations of 50 mg/L DDAC (MIC 250 mg/L) and 100 mg/L for ADBAC and all other QACs (MICs > 1,000 mg/L).¹⁷⁰ However, the study did not enable conclusions on whether complete mineralization occurred. In general, research pertaining to degradation of QACs in WWTPs is contradictory; most reports indicate that removal from wastewater is primarily based on sorption effects (not biodegradation), and that degradation of QACs by bacteria found in activated sludge is extremely limited.¹⁶⁹

Aerobic bacteria rapidly mineralize glutaraldehyde with glutaric acid as an intermediate. In both wastewater and seawater, aerobic biodegradation has been reported at concentrations of up to 50 mg/L.¹²² Under anaerobic conditions over a time period of 123 days, it was reported that glutaraldehyde biodegradation lead to the accumulation of 1,5-pentanediol and 3-formyl-6-hydroxy-2-cyclohexene-1-propanal, but no mineralization to CO₂ was observed.¹²²

For most of the other biocides, information on their biodegradation behavior is sparse and often only available through their reregistration eligibility decision reports without citation of peer-reviewed studies. DBNPA was reported to undergo aerobic and anaerobic biodegradation

on the order of hours, yielding six different products: oxalic acid, 2-cyanoacetamide, bromoacetamide, dibromoacetic acid, bromoacetic acid, and dibromoacetonitrile.⁸⁴ No information on complete mineralization was given. Biodegradation studies also showed that THPS is completely converted into CO₂ by bacteria.¹⁵⁹

Downhole conditions. As stated above, very little is known about the reactivity of chemicals under the high temperatures and pressures of downhole conditions. Previous research on produced water revealed that it did not contain all of the same organic compounds which had initially been injected into the well, suggesting that sorption and/or degradation had occurred underground.⁸⁹ As temperatures and pressures increase underground, chemical equilibrium will shift in accordance with Le Chatelier's principle, and reaction equilibrium will shift to favor endothermic products (negative Δ_rH) and products with smaller volume (fewer molecules and liquid/solid phase) as compared to standard state conditions (25 °C at atmospheric pressure). The high downhole pressures and temperatures may not only lead to unexpected chemical reactions or degradation, but may also alter the potential for biodegradation of organics (including biocides) underground.

The stability of aqueous organic chemicals at elevated temperatures is also highly influenced by the inorganic minerals present.¹⁷¹⁻¹⁷³ Therefore, it is important to attempt to understand the subsurface biogeochemistry that influences the fate of biocides¹⁷³ and other organic additives during hydraulic fracturing. Complete fracturing of a production well typically requires 3-5 days¹⁷⁴, and during this time period the inorganics in solution may either complex with or catalyze degradation of the organic additives in fracturing fluids. Furthermore, the extreme pressures utilized in hydraulic fracturing may work in conjuncture with elevated temperatures underground to produce chemical reactions unexpected under normal surface

conditions.^{171,175-177} Although research is currently ongoing in this area,^{87,88} the fate of biocides under these unique conditions is still unclear.

Toxicity

Most of the biocides used in fracturing fluids are severe eye and skin irritants, but have relatively low acute toxicity to mammals. Apart from a few exceptions, their oral median lethal dose (LD₅₀) values for rats range between 200 and >1,000 mg/kg (Table 3). However, the same biocides tend to be acutely toxic to aquatic life at low concentrations, especially to Mollusca such as oysters, which are highly sensitive to pollutants.¹⁷⁸ For example, glutaraldehyde displays only low to moderate toxicity towards aqueous organisms with the exception of oysters (LC₅₀ = 0.78 mg/L in the eastern oyster, *Crassostrea virginica*).¹⁷⁹ DBNPA is quite toxic to all aquatic life; the lowest reported LC₅₀ for *Daphnia magna* is 0.66 mg/L (48 h) and 1.0 mg/L for rainbow trout (96 h).^{84,180} Shellfish such as the estuarine oyster are especially sensitive to the presence of DBNPA, showing acute toxicity at levels below 0.070 mg/L.⁸⁴ Additionally, DBNPA has been shown to affect the reproduction of *D. magna* at the sub-toxic levels of 0.053 mg/L, and to negatively affect the growth of juvenile rainbow trout at concentrations as low as 0.019 mg/L¹⁸¹ and fathead minnow at 1.8 mg/L.¹⁸² DBA (breakdown product of DBNPA) also exhibits toxicity to aquatic life at very low concentrations¹⁸² (fathead minnow LC₅₀ is 0.55 mg/L within 96 h).¹⁸³ Bronopol is very toxic to marine invertebrates such as oysters – observed LC₅₀ is 1.6 mg/L for *D. magna*¹⁸⁴ and 0.77 mg/L for the Eastern Oyster.¹⁵⁴ It can therefore be concluded that substantial spills into surface waters or streams may have noticeable ecotoxicological effects on aquatic species.

Despite not being highly acutely toxic, certain biocides are suspected to possess developmental toxicity, carcinogenicity, mutagenicity, genotoxicity, and/or chronic toxicity (Table 3). Only a few of the hydraulic fracturing biocides have thus far been evaluated by the International Agency for Research on Cancer (IARC) or the U.S. EPA. For the remaining biocides, the evidence that does exist is insufficient to draw any firm conclusions.

Table 3: Reported toxicological data of biocides used in hydraulic fracturing operations. Lowest reported value is displayed. Active ingredient (AI) concentration is 100% unless otherwise stated. †Values normalized to 100% AI from reported values which use 50% AI. *Indicates mouse animal model.

Biocide	Mammalian (rat)		Chronic Toxicity
	LD ₅₀ Oral (mg/kg)	LC ₅₀ inhaled (4h, mg/L)	
ADBAC	305	0.054 – 0.51 ¹³³	No evidence found. ¹³³
Bronopol	325	0.588 ¹³⁷	Reproductive toxicity, degradation products formaldehyde is known human carcinogen (IARC 1) and nitrosamines are likely human carcinogens (IARC 2A). ¹³⁸
Chlorine dioxide	316	0.290 ²²¹	Developmental toxicity in lab animals. ²²¹
CMIT	105	0.330 ²²³	<i>Assumed to be same or similar to methylisothiazolinone (MIT).</i> ²²⁴
Dazomet	519	8.4 ¹⁴⁵	Developmental toxicity in lab animals: skeletal variations. ¹⁴⁷
DBNPA	207	0.320 ¹⁷²	Degradation product DBA is possible human carcinogen (IARC 2B) and developmentally toxic in lab animals. ^{169, 170}
DDAC	238	0.07 ¹³²	Developmental toxicity: skeletal abnormalities. No evidence of carcinogenicity. ¹³²
DMO	1,173	1.10 ²²⁷	Degradation product formaldehyde is known human carcinogen (IARC 1). ¹³⁸
Glutaraldehyde	460 [†]	>4.16 [†] ¹⁶²	No evidence found. ¹⁶²
Hypochlorite	5,800	no data ²²¹	Mutagenic <i>in vitro</i> . May react to form trihalomethanes, possible human carcinogens (IARC 2B). ²²¹
MIT	105	0.330 ²²³	Neurotoxic. No evidence of carcinogenicity. ²²⁴
N-Bromo	1,170	no data ²³⁰	No evidence found.
Peracetic acid	1,540	0.450 ²³²	No evidence found. ²³¹
THPS	290	0.591 ¹⁴²	Mutagenic <i>in vitro</i> , degradation product formaldehyde is known human carcinogen (IARC 1). ¹⁴²
TMO	1,173	1.10 ²²⁷	Degradation product formaldehyde is known human carcinogen (IARC 1). ¹³⁸
TTPC	1,002	<0.9 ¹⁶⁸	Developmental toxicity in lab animals. ¹⁶⁸

Though research is scarce, developmental toxicity (i.e., teratogenicity) has been observed from several of the lytic biocides used in fracturing fluids, such as DDAC and TTPC, as well as the conventional chlorine dioxide (Table 3). Observations of developmental toxicity associated

with DDAC have led to a recommended daily intake limit of 0.1 mg/kg/day for females of reproductive age.¹⁴⁹ While preliminary EPA studies suggest TTPC exhibits some developmental toxicity,¹⁸⁵ the inclusive final report has yet to be published. Other biocides have shown developmental toxicity associated only with very high concentrations, such as Bronopol,¹⁵⁴ THPS,¹⁸⁶ or *in vitro*, such as MIT.¹⁸⁷

Carcinogenicity is rare among the biocides used for fracturing fluids (Table 3), and often is confirmed only *in vitro* at very high concentrations. Typically, any carcinogenicity is due to the breakdown products of the biocides instead of the parent compound themselves, such as the possible human carcinogen DBA (IARC Group 2B), a breakdown product of DBNPA that has been shown to induce tumors in rat liver cells *in vitro*¹⁸⁸ and to have carcinogenic effects in lab animals when administered in concentrations over 100 mg/L.¹⁸⁹ Other examples include nitrosamines produced from hydrolysis of Bronopol,^{156–158} many of which are probably carcinogenic to humans (IARC Group 2A).^{165,190} Also, several of the biocides used in hydraulic fracturing operations are known or suspected formaldehyde generators. Formaldehyde is a known human carcinogen (IARC Group 1) if inhaled.^{154,155} However, when dissolved in aqueous media, formaldehyde exists predominantly in its less toxic hydrated form, methanediol (CH₂(OH)₂, methylene glycol), and its tendency to evaporate from aqueous solution is low.^{191,192} Examples of formaldehyde generators include Bronopol,¹⁵⁴ THPS,^{32,161,193,194} and Dazomet,^{32,162–164} however, none of these have been found to be carcinogenic in laboratory tests.¹⁹⁵

A few compounds (though not carcinogenic) have been found to mutate DNA *in vitro*. Examples of mutagenic biocides include glutaraldehyde^{196–198} and THPS.¹⁸⁶ Other biocides, though not producing mutations, have been found to exert genotoxic effects *in vitro*, such as ADBAC in plant root tip cells and human lymphocytes at concentrations of 1.0 mg/L.¹⁹⁹

However, *in vitro* effects do not necessarily imply human effects. Genotoxic effects from ADBAC exposure have also been observed in human respiratory cells (study used 1,000 mg/L²⁰⁰) and fish cells at 0.12 mg/L.²⁰¹ This study also revealed that DDAC (or its bromide salt, respectively) exerted genotoxicity towards mammalian cells at 0.3 mg/L.¹⁹⁹ Finally, the electrophilic biocide THPS (or its chloride salt, respectively) was reported to be genotoxic to fish cells at concentrations of 0.2 µg/L.²⁰¹

For a holistic risk assessment, the mobility and degradability of hydraulic fracturing biocides have to be included, which may limit their ability to exert toxicity on humans or other organisms. Attention must be paid, however, to transformation products that are known to be more toxic and /or more persistent than the parent biocide, such as DBNPA and its product DBA, further emphasizing the need for more detailed degradation studies. The charged, cationic biocides may be more resistant to breakdown, but their strong sorption to clay and organic matter is known to reduce their toxicity.²⁰²

Alternative Approaches

Alternative products and technologies with biocidal activity exist or are currently being explored to enable a pathway to more sustainable bacterial control. However, none of them are without any environmental and/or human health impacts. One biocide that has been recognized for its relatively low environmental impact is THPS, which received the Presidential Green Chemistry Award in 1997 due to its low toxicity, low treatment levels, and rapid breakdown when used properly.²⁰³ However, a surface spill may result in formaldehyde generation due to the evaporation-driven conversion from its hydrated form, methanediol, a breakdown product of THPS.

Peracetic acid is a biocide already used in hydraulic fracturing operations, although thus far only in 0.01% of all wells registered on FracFocus (Figure 3). It is a stronger oxidizing agent than H₂O₂, but has not been found to produce harmful DBPs²⁰⁴ and is not expected to have adverse health effects including carcinogenesis.²⁰⁵

Ozone has been widely used for drinking water and food disinfection.^{206–208} It readily reacts with double bonds in critical cellular components, but is known to generate some DBPs such as bromate.^{209,210} Also, health effects such as various respiratory diseases, heart attack, and premature death are linked with ozone and its associated pollutants.²¹¹ Furthermore, ozone's rapid reaction kinetics are disadvantageous for downhole applications, which usually take days to weeks to completion. As with other oxidizing agents including peracetic acid, it may induce metal corrosion and thus steel casing failure.

Chlorine dioxide has a longer lifetime than ozone and is thus already used in hydraulic fracturing fluids (Table 1). Its corrosion potential is comparably low, but it may produce carcinogenic DBPs.^{212,213} Because total dissolved organic carbon in produced water has been measured as high as 5,500 mg/L,^{13,75} formation of DBPs resulting from use of chlorine dioxide, chlorine (generated by sodium hypochlorite), and bromine (generated by N-bromo) must be considered.²¹⁴ A previous study correlated increased levels of THMs in WWTP effluent to Marcellus shale wastewater, especially brominated species, were observed.²¹⁵ Furthermore, chlorine dioxide gas is explosive when exceeding 10% v/v in air and must thus be produced on site. Historical illnesses outside hydraulic fracturing activity including one fatality resulting from exposure to less than 19 ppm have been reported.²¹⁶

The injection of nitrate or nitrite has been broadly used as an alternative electron acceptor for microbial respiration to avert unwanted sulfate reduction.^{217–221} Especially in offshore water

flooding operations, where seawater is continuously injected downhole, nitrate was proven to be very effective in reducing amount and activity of SRB.²²²

There are other alternatives which circumvent the need for chemical addition altogether, such as use of ultrasonic radiation²²³ or UV light.²²⁴ These technologies have proven to be extremely effective for microbial control, but their high energy demands and lack of residual effect currently prevent them from widespread use. Furthermore, electrochemical approaches, such as generation of electrochemically activated solutions,²²⁵ can be used to generate reactive oxygen species such as hydroxyl radicals. However, in the presence of high chloride concentrations, electrochemical techniques can also generate hypochlorous acid (HOCl), which, as stated above, is known to produce DBPs. Due to their typically much lower energy consumption, electrochemical technologies are widely applied in drinking water and wastewater treatment plants.²²⁶ Although their use for downhole applications is limited, they may be viable alternatives for hydraulic fracturing-related aboveground water operations.

Discussion and Future Work

Based on the currently available data regarding usage, mobility, degradation, and toxicity of the biocides used in hydraulic fracturing operations, the following conclusions can be drawn: (1) While the uncharged species can be mostly expected to be in the water phase where they are subject to biotic or abiotic degradation, the cationic quaternary ammonium and phosphonium compounds will strongly sorb to soil or sediments, where their (bio)availability to degrade may be limited and their toxicity attenuated. (2) In surface and shallow subsurface environments, many of the biocides are degradable through abiotic and biotic (especially aerobic) processes, but some can potentially transform into more toxic or persistent compounds, which may accumulate

under certain conditions. (3) Although being a critical process, the understanding of the biocides' degradation and sorption under downhole conditions (high pressure, temperature, salt and organic matter concentrations) is extremely limited, currently not enabling reliable risk assessment. (4) Efforts in developing alternative biocides with lower human health risks are ongoing. Application of existing alternatives is currently limited by high cost, high energy demands, inapplicability under downhole conditions, or formation of DBPs.

Critical data regarding the fate and transport of hydraulic fracturing biocides are still missing despite the widespread and long-term use in other water-processing industries. This is especially true for biotic and abiotic (e.g., photolytic, hydrolytic) degradation processes, which are key to controlling organic contaminant removal from the environment. Future research addressing biocide degradation rates should thus focus on fundamental laboratory- and field-scale investigations under various environmental conditions as well as their runoff and leaching potential. Furthermore, as noted above, due to the unique exposure to deep geological formations either during hydraulic fracturing or after injection into deep disposal wells, their reactivity and sorption behavior downhole needs to be considered.

The biocides' susceptibility to various transformation²²⁷ and degradation processes in combination with the fact that some degradation products have been reported to be more toxic and/or persistent than the parent compound highlight the need for appropriate analytical detection methods to more accurately assess their potential environmental impact. Excellent screening methods based on high-resolution mass spectrometry exist for polar organic molecules^{228–230} and breakdown products,^{227,230–232} which could theoretically be modified and applied towards detecting hydraulic fracturing chemicals. Modification (typically in sample preparation) is necessary to accommodate the high salinity and TDS commonly associated with

fracturing wastewater or measurement accuracy may suffer due to matrix suppression effects, as was recently found responsible for underestimation of radium levels in produced water from the Marcellus shale.²³³

Furthermore, future environmental fate and transport studies need to address the complexity of hydraulic fracturing fluids. The presence of other organic additives, especially polymers that increase fluid viscosity like gelling agents or the friction reducer polyacrylamide,⁹⁷ may have substantial impacts on their transport through soil. Vice versa, the presence of biocides above their minimum inhibitory concentrations may impact natural biodegradation processes of other organic fracturing fluid additives.

As of today, no groundwater contamination by hydraulic fracturing additives in the U.S. has been irrefutably documented in the peer-reviewed literature.^{75,97,121} However, due to the fact that hydraulic fracturing fluid spills have been reported previously (e.g., there were 591 documented spills in 2013 in Colorado alone),⁹⁶ this scenario does not appear unlikely. Consequently, it appears mandatory to address this concern at a minimum through (1) adequate baseline monitoring in combination with (2) comprehensive chemical analyses beyond currently analyzed and suggested chemical reporting parameters (i.e. bulk water characteristics like pH and TDS, and concentration of inorganics including chloride, sodium, calcium, barium, strontium, magnesium, radium, uranium, and iron).²³⁴ In case of inadvertent releases, these data will enable the development of appropriate cleanup strategies, allow for ecosystem and human health risk assessment, and thus pave the way for a more sustainable approach to natural resource extraction.

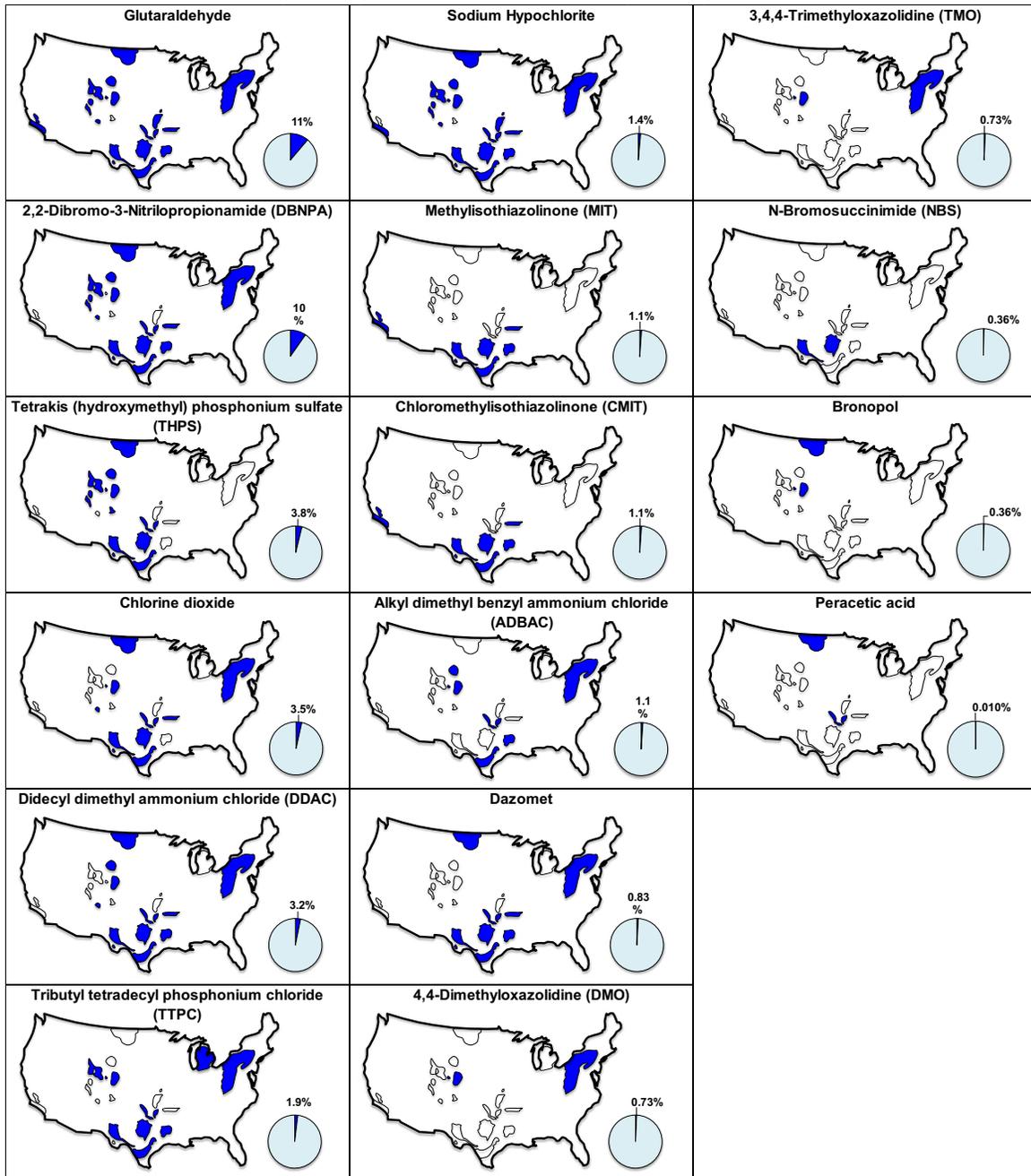


Figure 4. U.S. shale plays (in blue) and percentage in which the respective biocides have been used, normalized to the total number of wells (62,887 registered), including wells fractured without biocide use. Data collected from FracFocus¹ is pertinent to wells fractured after January 01, 2011. Count includes all precursor compounds where applicable.

Table 4. Values for octanol-water partition constant (K_{OW}), water solubility, and soil organic carbon-water partition constant (K_{OC}) estimated via EPI Suite v4.11,² ALOGPS 2.1,³ ChemAxon,⁴ and XLOGP2,⁵ and comparison to reported experimental values where available. Values in italics indicate that missing parameters in the database are preventing accurate calculation of this value (as reported by given calculation software).

Biocide	log K_{ow}					Water sol. @25°C (mg/L)			log K_{oc}		
	Exp.	EPI Suite	ALOGPS	XLOGP2	Chem Axon	Experimental	EPI Suite	ALOGPS	Exp.	EPI Suite (Kow)	EPI Suite (MCI)
Alkyl dimethyl benzyl ammonium chloride (ADBAC)	no data	3.91	1.71	6.34	1.74	>350,000 ⁹	2.203	<i>0.00566</i>	5.810 ¹⁰	3.001	5.955
Bronopol	no data	-0.64	-0.21	-0.70	-0.20	200,000 ¹¹	62,850	12,830	no data	0.033	0
Chloro-methylisothiazolinone (CMIT)	no data	-0.34	0.85	0.72	1.35	no data	148,700	50,020	no data	0.711	1.287
Dazomet	0.63 ¹⁴	0.94	0.48	-0.02	1.28	3,000 ¹⁴	19,360	7,080	1.00 ¹⁴	2.44	1.923
Dibromo-nitripropionamide (DBNPA)	0.82 ¹⁴	1.01	0.64	0.79	0.24	15,000 ¹⁴	2841	3,290	no data	1.767	0.731
Didecyl dimethyl ammonium chloride (DDAC)	≈0 ¹⁴	4.66	4.54	8.26	4.01	700 ¹⁹	0.5505	<i>0.00391</i>	5.64-6.20 ¹⁸	3.416	5.686
Dimethyloxazolidine (DMO)	0.73 ²¹	-0.08	-0.20	0.44	0.40	miscible ²⁰	miscible	470,000	no data	0.747	0.975
Glutaraldehyde	-0.33 ²⁴	-0.18	0.93	0.48	-0.27	miscible ²²	167,200	64,030	2.07-2.70 ²⁴	0.367	0
Methylisothiazolinone (MIT)	no data	-0.83	-0.09	0.48	0.23	no data	536,700	130,000	no data	0.44	1.082
N-Bromosuccinimide	no data	-1.19	-0.65	0.05	-0.08	14,700 ¹⁴	581,200	140,000	no data	0.259	0.481
Peracetic acid	no data	-1.07	-0.70	-0.58	-0.30	miscible ²⁶	miscible	360,000	no data	0.104	0.179
Tetrakis hydroxymethyl phosphonium sulfate (THPS)	no data	-9.77	-0.87	-6.52	-3.49	400,000 ¹⁴	miscible	9,870	no data	-5.302	1
Tributyl tetradecyl phosphonium chloride (TTPC)	no data	6.48	8.17	11.53	10.02	miscible ²⁹	0.00538	<i>0.00334</i>	no data	5.624	7.66
Trimethyloxazolidine (TMO)	no data	0.13	0.38	0.68	0.78	miscible ²⁰	821,900	700,000	no data	0.841	0.981

Table 5. Average absolute errors of experimental vs. calculated values for octanol-water partition constant (K_{ow}) and soil organic carbon-water partition constant (K_{oc}) for compounds with a known experimental value. Only absolute values used in error analysis.

	Lytic Biocides	Electrophilic Biocides	All
EPI Suite	4.66	0.252	0.882
ALOGPS	4.54	1.30	1.71
XLOGP2	8.26	0.509	1.62
ChemAxon	4.01	0.509	1.01
	Av. absolute error in log Koc		
	Lytic Biocides	Electrophilic Biocides	All
EPI Suite Kow Method	2.66	1.73	2.19
EPI Suite MCI Method	0.190	4.96	2.57

CHAPTER 3 – DOWNHOLE TRANSFORMATION OF THE BIOCIDES

GLUTARALDEHYDE⁸⁷

Introduction

Because of the variability inherent to downhole conditions between different wells, HFF additives are exposed to a wide variety of underground environments. In order to explore the potential impact of downhole conditions on organic HFF additives, the biocide glutaraldehyde was selected as a representative chemical because it belongs to an important group of toxic chemicals routinely added to HFFs. Biocides are used for underground microbial control to prevent the detrimental side-effects of bacterial growth and anaerobic respiration: production of toxic H₂S gas (thus “souring” the natural gas/oil and posing a health risk to workers) and corrosion of underground casing.²⁸ Because GA exercises its biocidal activity by cross-linking primary amines contained within proteins on bacterial membranes, it is an effective and non-specific killer of microbiota.^{28,152} As the most commonly used biocide, it can therefore be assumed that GA is also exposed to the widest range of deep subsurface conditions, making a natural choice of test chemical.

Spills of HFFs and use of treated oil and gas wastewater for crop irrigation have raised public concern about risks to water and food safety.^{42,235} Unfortunately, little is known about the toxic chemicals returning to the surface after hydraulic fracturing, , as so little is known about the transformation and fate of these chemicals downhole. Therefore, using GA as an example, the manner in which extreme conditions within deep shale reservoirs can change HFF composition is demonstrated. With stainless steel reaction vessels imitating well casing, laboratory experiments simulating the aspects of hydraulic fracturing will be performed, during which the concentration

of GA will be monitored. The main downhole environmental stressors— namely temperature, pH, pressure, salinity, and presence of shale— will be tested for effects on the kinetics of removal of GA from solution. Ultimately, chemical analysis will be performed to deduce the transformation products of GA thereby lending insight to the final chemical products which may be present in flowback and produced water.

Experimental Section

Reaction Vessels. Deep subsurface (downhole) conditions were simulated in 600-mL capacity stainless steel high pressure/temperature vessels (model no. 4768, MAWP 3,000 psi/200 bar at 350 °C) (Parr Instrument Company, Moline, IL) outfitted with four ports: one for gas, one for liquid, one for a thermocouple, and one for a pressure gauge. Stainless steel reactors were chosen to simulate the potential interactions that may occur between the well casing and the injected chemicals. The thermocouple was connected to a dual-channel temperature control unit (J-KEM Scientific, Saint Louis, MO), which regulated internal temperature via heavy insulated flexible heat tape (BriskHeat, Columbus, OH) wrapped around the exterior of the vessels. A stir bar was added to each vessel to ensure even heating of internal solutions. Pressurized N₂ atmosphere was controlled using the reactors' pressure gauges (Swagelok, Solon, OH) and fed to the vessels through high-pressure threaded hosing and valves (Swagelok, Solon, OH) attached to an N₂ gas cylinder (99.9% purity) outfitted with a high-pressure regulator (Airgas, Radnor Township, PA). This setup is capable of heating solutions to 350°C and pressures up to 20.7 MPa.

Experimental Solutions. All chemicals were of ACS grade (Sigma-Aldrich, St. Louis, MO) unless otherwise stated. GA solutions were prepared in 18 MΩ DI H₂O (Millipore, Billerica, Massachusetts) at a concentration of approximately 5.0 mM, matching a working 500

mg/L concentration. Control of pH was achieved by addition of 25 mM phosphate buffer, slightly exceeding a 5:1 molar ratio to GA. Synthetic brine solutions consisted of phosphate buffer and NaCl (made to concentrations of 1 M, 2 M, and 2.8 M). All reaction vessels and solutions were purged immediately before each run with N₂ gas using a separate purging flask equipped with a glass frit. After transfer of experimental solution from flask to vessels using N₂ gas, the setup was reconnected and carefully pressurized while brought to target temperature. Time zero is marked when temperature comes within 1 °C of that desired (matching the error of the temperature control capabilities of the setup). Samples are then collected at regular time intervals over the length of each experiment.

Quantification of Glutaraldehyde (GA). Samples were allowed to cool to ambient temperature (23±1 °C) as monitored by an IR thermometer before derivatization in triplicate as follows: a 100-μL aliquot of analyte-containing solution was added to 900 μL of 0.121 M HCl and 3 mL of 2,4-dinitrophenylhydrazine (DNPH) solution (0.0101M in acetonitrile, HPLC grade). Derivatization was performed both to consolidate hydrated forms of GA in order to idealize chromatographic separation, as well for the addition of a chromophore for improvement of detection and quantification. GA was quantified via an Agilent 1200 series HPLC system equipped with a UV-diode array detector (HPLC-UV; Agilent Technologies, Inc., Santa Clara, CA) using a detection wavelength of 358 nm. The chromatographic method used a Zorbax Eclipse Plus C18 column (150 x 4.6 mm i.d., 5 μm particle size, Agilent Technologies, Inc., Santa Clara, CA) maintained at 40 °C, a constant flow rate of 3.0 mL/min, and an isocratic mobile phase of 70:30 acetonitrile:water (both +1% formic acid) with 10 μL sample injection volume. GA elutes as two peaks (representing each dominant structural isomer) at 1.9 and 2.1

minutes, both of which are integrated for quantitation. External calibration curves were utilized for quantitation.

Electrospray Ionization Time-of-Flight Mass Spectrometry (ESI-TOF-MS). Mass spectra were recorded with an HPLC (1200 model, Agilent Technologies, Inc.) and time-of-flight mass spectrometer (TOF-MS) (6220 model, Agilent Technologies, Inc.). The collected fractions were re-eluted from a C18PFP 2.1x150 mm with 3.0 μm particle size column (Phenomenex, Torrance, CA). The instrumental conditions were as follows: capillary voltage was 2,000 V, skimmer voltage was 60.0 V. Cone gas was N_2 , held at 310 $^\circ\text{C}$, 8.0 L/min, with a nebulizer pressure of 310 kPa (45 psi). Data collected between a mass range of 118-3,200 with a scan rate of 1.05. All mass spectra collected in positive ion mode.

Shale Sample. Shale samples used in this experiment were collected from an undisclosed site within the Marcellus Formation and provided by Multi-Chem, a Halliburton Service. The average BET specific surface area was determined to be 15.0 m^2/g , the total carbon content was 9.1 %. Samples were stored in glass and refrigerated (approx. 5 $^\circ\text{C}$). Shale samples were dried in a desiccator for 48 hours prior to weighing and subsequent use in experiments. The reaction vessels including the shale samples were then purged to minimize exposure to O_2 by filling and draining the vessel with N_2 three times, then holding the vessel at pressure under $\text{N}_{2(\text{g})}$ overnight prior to running an experiment.

Kinetic studies. Given a system with the reactants GA and hydroxide (OH^-), the rate order may be written as follows:

$$\text{Rate} = -\frac{1}{x} \frac{d[\text{GA}]}{dt} = k[\text{GA}]^x [\text{OH}^-]^y$$

Isolation of a single chemical species to determine its rate order may be achieved by a method akin to flooding the system with one of the reactants such that its concentration is constant with

respect to time; in this case, OH^- was held constant through use of a buffer. With this technique, the rate constant k may be replaced by the pseudo-rate constant k' and the rate law re-written as:

$$\text{Rate} = k'[\text{GA}]^x$$

Where $k' = k[\text{OH}^-]^y$, and $[\text{OH}^-]$ is held constant.

To test whether pseudo- first or second order kinetics were being exhibited, the linearity of $\ln[\text{GA}]$ vs. t (to test for first order) was compared to the linearity of $1/[\text{GA}]$ vs. t (to test for second order), as per the following rearrangements of the first- and second-order rate law, respectively:

$$\ln[\text{GA}] = -k't + \ln [\text{GA}]_0 \quad \text{and} \quad \frac{1}{[\text{GA}]} = k't + \frac{1}{[\text{GA}]_0}$$

When data is graphed using these equations, the model providing the better linear regression (closest to 1) or fit indicates the reaction order of GA. Note that only values gathered prior to the first half-life were used in linear regression models. Order dependence of OH^- was determined by writing the expression for k' in the form of a straight line and using it to graph data from three experiments with varied concentrations of OH^- where all other parameters (such as T) are held constant. In this form, the slope of the resulting line can be used to solve for y , the rate order of $[\text{OH}^-]$.

$$\ln k' = y \ln[\text{OH}^-] + \ln k$$

Half-lives ($t_{1/2}$) were generated by using $[\text{GA}] = 1/2[\text{GA}]_0$ to generate the following equation, after which insertion of experimentally determined k' values along with $[\text{A}]_0$ enabled $t_{1/2}$ determination.

$$t_{1/2} = \frac{1}{k'[\text{GA}]_0}$$

The Arrhenius equation was used to determine the frequency factor A and the activation energy E_a for use in the kinetic model:

$$k = Ae^{\frac{-E_a}{RT}}$$

where T is temperature in Kelvin, and R is the universal gas constant. By taking the natural log of both sides followed by rearrangement, this equation may be written in the form of a straight line:

$$\ln k = -\frac{E_a}{R} * \frac{1}{T} + \ln A$$

For each temperature experiment (with a constant pH of 7), $\ln(k)$ was plotted versus $1/T$ to generate a straight line ($R^2 = 0.99976$). The slope of this line was equated to $-E_a/R$ to determine E_a , and the y-intercept was equated to $\ln(A)$ to determine A.

Results and Discussion

Impact of Deep Subsurface Matrix Parameters. To delineate the impact of deep subsurface geophysical and chemical stressors (“matrix parameters”) on the fate of HFF additives, the transformation kinetics and pathways of the proof-of-concept chemical GA were systematically evaluated within stainless steel reactors in the laboratory. The most relevant matrix parameters inducing chemical stress under downhole conditions (and thus the parameters tested here) were decided to be temperature, pressure, pH, salinity (commonly analyzed as total dissolved solids, TDS), and absence/presence of shale (the latter enabling discrimination between bulk water and shale surface processes such as sorption). The reference conditions chosen for a basis of comparison for GA transformation were 100 °C, 690 kPa, pH 7, ≈ 0 M NaCl, and 0 g shale. The concentration of GA was measured over time as matrix parameters were initially varied individually; the initial volume of the reactors was 500 mL and sample volumes were approximately 5 mL each. The following parameters including reference

conditions (italicized) were tested: 65, *100*, and 140°C; *690* and 11,000 kPa; pH 6, *pH* 7, and pH 8; 0, 1, 2, and 2.8 M [NaCl]_{aq}; and 0, 12, 24, and 36 g of shale added to the reactors.

The results shown in Figure 5 indicate that within the parameter range in which GA is used in HFF, temperature has a large effect on GA removal from the (bulk) aqueous phase with transformation rate increasing dramatically as temperature rises. The second very influential parameter on GA removal is pH, showing a pronounced enhancement of transformation kinetics as conditions become more basic (again within environmentally relevant ranges). Removal of GA from aqueous solutions via formation of insoluble polymers or protein cross-linking has been reported previously under normal atmospheric conditions,¹⁵² but never before verified in a simulated underground environment.

However, the impact of salinity on the stability and transformation of GA has never before been investigated or reported in the literature, and remains an important matrix parameter potentially affecting the downhole behavior of GA (see Figure 4). A parameter often overlooked as having an insignificant effect on transformation kinetics, it is observed here for the first time that NaCl concentrations matching those found in real-world produced water samples^{8,10,15,24,236} significantly alter the rate of GA transformation kinetics. In fact, it was found that with each 1 M NaCl increase, the observed transformation half-life ($t_{1/2 \text{ obs}}$) also increased by a factor of two: furthermore, the relationship between [NaCl]_{aq} and $1/k_{\text{obs}}$ (and therefore the relationship between [NaCl] and $t_{1/2}$) was highly linearly correlated ($R^2 = 0.9946$) (Figure 2), indicating that a salt concentration-based phenomenon is somehow hindering the transformation mechanism of GA; this will be discussed in further detail below. This effect was verified at one separate temperature (140°C with [NaCl] = 2M, Table 7.) to confirm the kinetic alterations are indeed additive.

In contrast, both pressure and increasing amounts of shale had no observable effect on GA removal (Figure 5), indicating that the presence of shale did not result in significant sorption or catalysis. However, it is noted that the experimental setup was limited and the full range of conditions representative of unconventional reservoirs was not tested for these two parameters; pressure in some instances can exceed 100,000 kPa, while actual fluid-to-shale ratios are closer to 5:95.¹⁰²

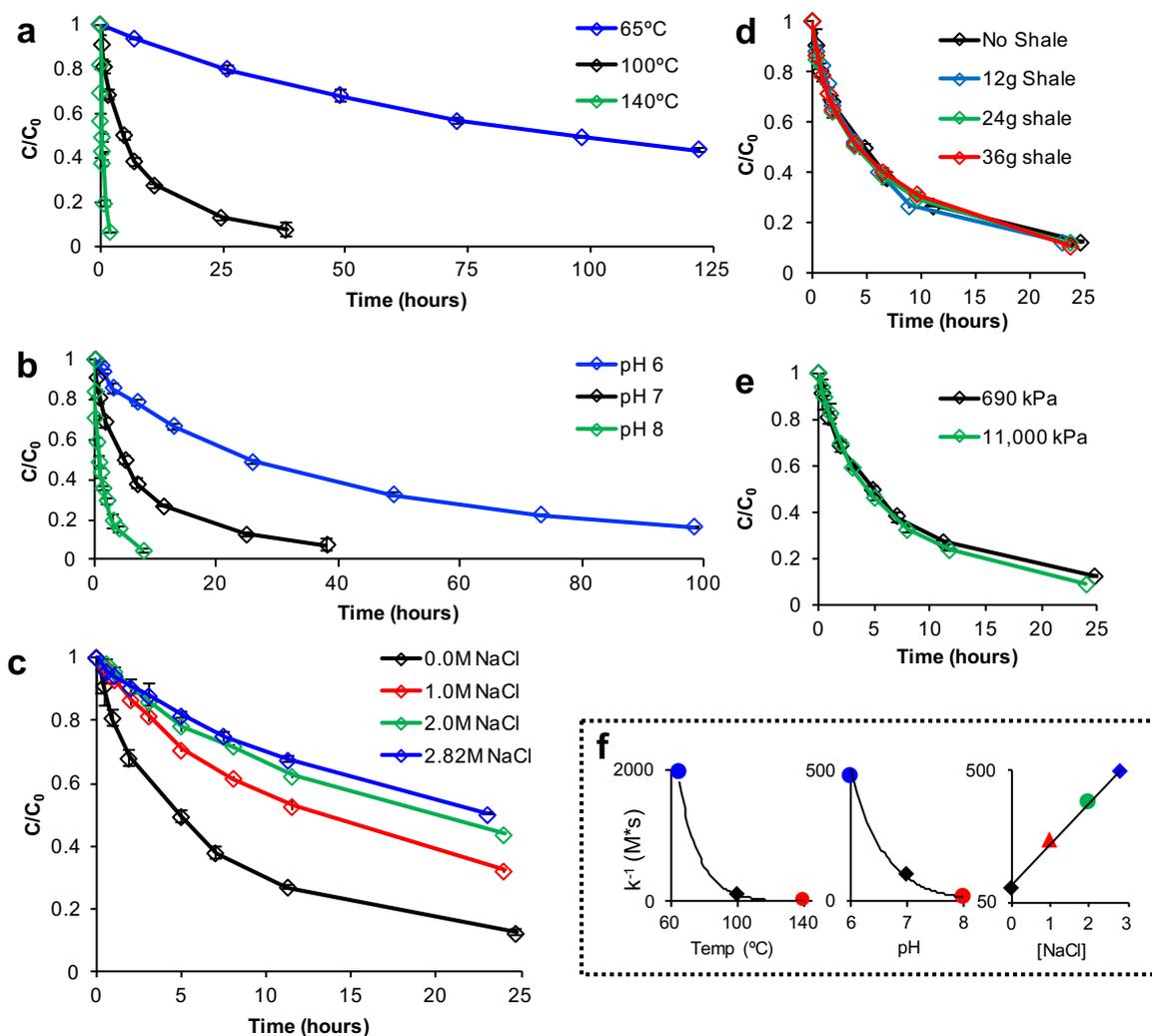


Figure 5. Transformation kinetics of glutaraldehyde (GA; $C_0 = 500$ mg/L) under simulated downhole conditions. Panels a-e represent the degradation of GA (C/C_0) over experimental time in hours while modulating underground parameters a) temperature (T), b) pH, c) salinity as [NaCl], d) presence of shale, and e) pressure (P). Panel f displays the highly predictable manner in which the calculated rate constant k varies with the given parameter. The baseline conditions (100 °C, pH 7, 690 bar, 0 M NaCl) were used for these experiments while varying one variable. While T, pH, and [NaCl] (a-c) show significant dependence on the varied parameter, shale and P (d and e) do not affect the transformation kinetics of GA. Error bars represent error in chemical measurement, displaying 95% confidence intervals. Data in a-d were collected in the absence of shale and are thus representative of bulk aqueous processes.

Transformation Pathways, Kinetics, and Mechanisms. At the end of the reactor-based experiments (post-run), transformation products were both analytically detected in the aqueous solution (Table 6) and observed visually as a brownish black, highly viscous solid phase adhered

to the inside of the reactor (Figure 6). Accurate mass spectrometry revealed the major aqueous-phase transformation products to be a GA dimer and both doubly-hydrated and triply-hydrated isomeric trimers, in perfect agreement with previous chemical analyses of real-world flowback and produced water from Colorado, thus validating these experimental simulations.^{145,236,236} Full mass balance was not possible due to the strong adherence of the polymer precipitates to the stainless steel reactor walls.

Table 6. Putative identifications of dissolved GA transformation products based on data gathered via time-of-flight mass spectrometry.

Retention Time (min)	Measured Mass (m/z)	Base Peak Formula	Base Peak	Putative Identification	Theoretical m/z	Error (ppm)
6.6, 6.8	165.0931	$C_{10}H_{12}O_2H_+$	$[M+H]^+ - H_2O$	GA dimer $C_{10}H_{14}O_3$	165.0916	12.3
11.2	318.1913	$C_{15}H_{24}O_6N_4H_4^+$	$[M+NH_4]^+ + 2H_2O$	GA trimer $C_{15}H_{20}O_4$	318.1917	0.51

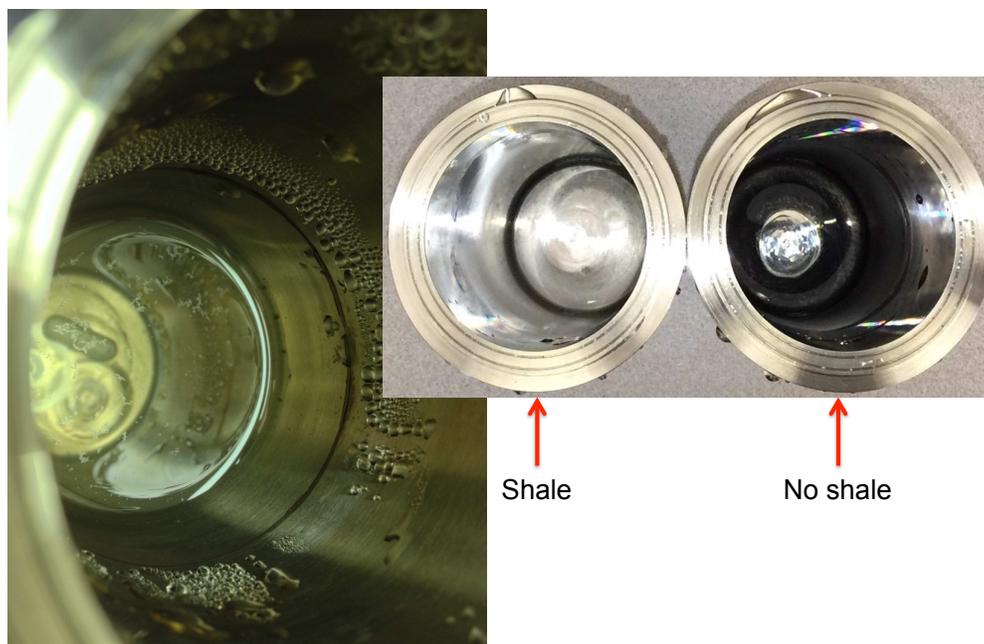


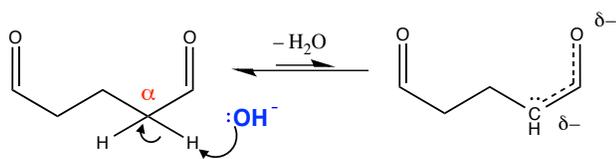
Figure 6. Pictures of the reaction vessel post-experiment, with and without shale.

Table 7. Summarized kinetic data for glutaraldehyde collected from reactor experiments without shale, including experimental conditions (temperature, pressure, pH, [NaCl], [GA]_{0 exp}) as shaded in red, calculated GA transformation rate constants and half-lives as shaded in green, and modeled results plus their error (relative deviation from t_{1/2 obs}) as shaded in blue. Note that the first row, italicized, represents baseline conditions and only changes from baseline conditions are indicated in the rows below. Each row of data represents an individual run, while the error in row 1 for *k'*_{obs} is the standard deviation between experimental replicates (n = 6).

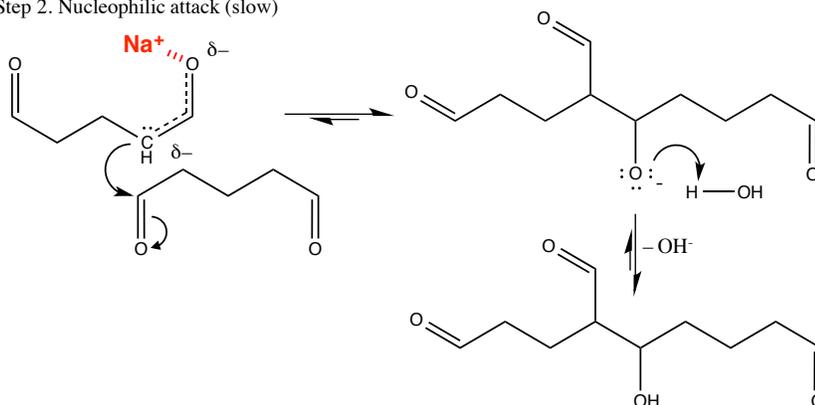
Temp (°C)	pH	P (kPa)	[NaCl] (M)	[GA] _{0 exp} (mg/L)	<i>k'</i> _{obs} (M ⁻¹ s ⁻¹)	t _{1/2 obs} (h)	t _{1/2 model} (h)	t _{1/2 model} % error
100	7	690	0	558.9	2,830 (±190)	4.89	4.64	-5.11%
65	–	–	–	568.1	124	94.1	99.5	5.73%
140	–	–	–	454.5	48,400	0.293	0.320	9.22%
–	6	–	–	564.2	3,010	23.7	27.1	14.3%
–	8	–	–	536.3	2,970	0.729	0.822	12.8%
–	–	11,000	–	577.4	2,730	4.33	4.64	7.16%
–	–	–	1	570.8	937	12.8	11.8	-7.81%
–	–	–	2	566.4	624	19.3	19.1	-1.04%
–	–	–	2.8	582.3	490	23.8	24.3	2.10%
140	–	–	2	510.8	10,900	1.23	1.19	-3.25%

The identified dissolved transformation products match an aldol condensation pathway by which GA polymerizes with another GA molecule (Figure 7). Mass spectrometric identification of the produced viscous products was not performed due to their insolubility in both water and organic solvent (perhaps due to strong sorption to the stainless steel walls, Figure 6), which may indicate precipitation from the aqueous phase due to molecular size rather than hydrophobicity.²³⁷ Based on both these laboratory observations and prior field-scale results²¹ it is reasonable to conclude that successive aldol condensations produce increasingly larger polymers which eventually precipitate out of solution. It is noted that experiments including shale resulted in preferential sorption of insoluble precipitates to the shale's hydrophobic surface rather than the stainless steel reaction vessel (Figure 6).

Step 1. Formation of enolate ion (slow)



Step 2. Nucleophilic attack (slow)



Step 3. Base-catalyzed dehydration (fast)

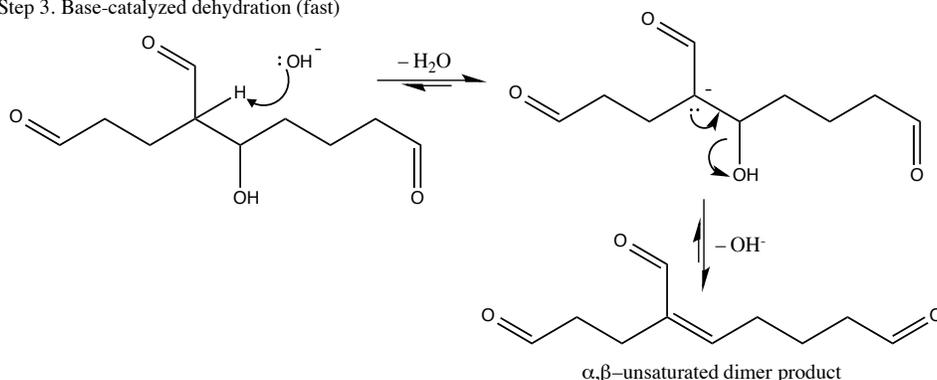


Figure 7. The reaction mechanism describing the aldol condensation between two monomer units of GA. Note the two important potential effects of salt on the chemical mechanism, represented by blue (OH^- in step 1) and red (Na^+ in step 2). Addition of salt may either: 1) depress available OH^- by increasing dissociation of weak acids thus lowering the pH and slowing step 1, or 2) Na^+ and the GA enolate ion could be participating in ion pairing thus slowing step 2. This reaction proceeds onward between GA subunits to create GA polymers of increasing size.

Fitting the experimental data at various fixed pH values yields correlation values of 0.994

$\geq R^2 \geq 0.999$ in all cases when matched to the linearized second-order rate equation:

$$\frac{1}{[\text{GA}]} = k't + \frac{1}{[\text{GA}]_0}$$

where [GA] is the aqueous concentration of the GA monomer (in M), k' is the pseudo-second-order rate constant (in $M^{-1}s^{-1}$), t is time (in s), and $[GA]_0$ is initial GA concentration (in M). This linear relationship verifies that the reaction kinetics are pseudo-second-order, further supporting the conclusion of GA autopolymerization (which is known to follow second-order kinetics¹⁵²). The overall rate equation was determined to be:

$$\text{Rate} = -\frac{1}{2} \frac{d[GA]}{dt} = k[GA]^2[OH^-]^{0.77}$$

where k is the overall rate constant (in units of $M^{-1.77}s^{-1}$). The rate order for $[OH^-]$ (accounting for the pH-dependency of the polymerization reaction) was determined to be between zero- and first-order kinetics at 0.77. These two rate orders indicate that both steps 1 and 2 of the aldol condensation mechanism (Figure 7) are partially rate-controlling, which has been previously observed in the analogous aldol condensation of acetaldehyde.²³⁸ Based on these rate orders, observed pseudo-second-order rate constants (k'_{obs}) and calculated half-lives ($t_{1/2 \text{ obs}}$) were determined for GA (Table 7).

Characterization of Transformation Products. During mass spectrometry of the transformation products, it is interesting to note that the dimer fragment $m/z = 165$ was associated with all chromatographic peaks despite the soft ionization scheme (see Figure 8 for proposed fragmentation explanation). This fragmentation is most likely an artifact of ionization, and is strong evidence that all products formed are polymeric relatives.

As with aqueous glutaraldehyde, we suspect that these compounds undergo repeated hydration/cyclic hemiacetal formation as shown in Figure 8. This could explain the lack of characteristic conjugated aldehyde absorbance near 310 nm in the UV-vis spectrum of the dimer isomers,²³⁹ while it is seen in the UV-vis spectrum of the trimer (identified later via mass spec, Table 6). The strong absorbance of the dimer near 230 nm may most likely be attributed to the

presence of the conjugated double bonds;²³⁹ due to the conjugation of these chromophores, absorbance is red-shifted 30 nm according to the Woodward-Fieser rules.²³⁹

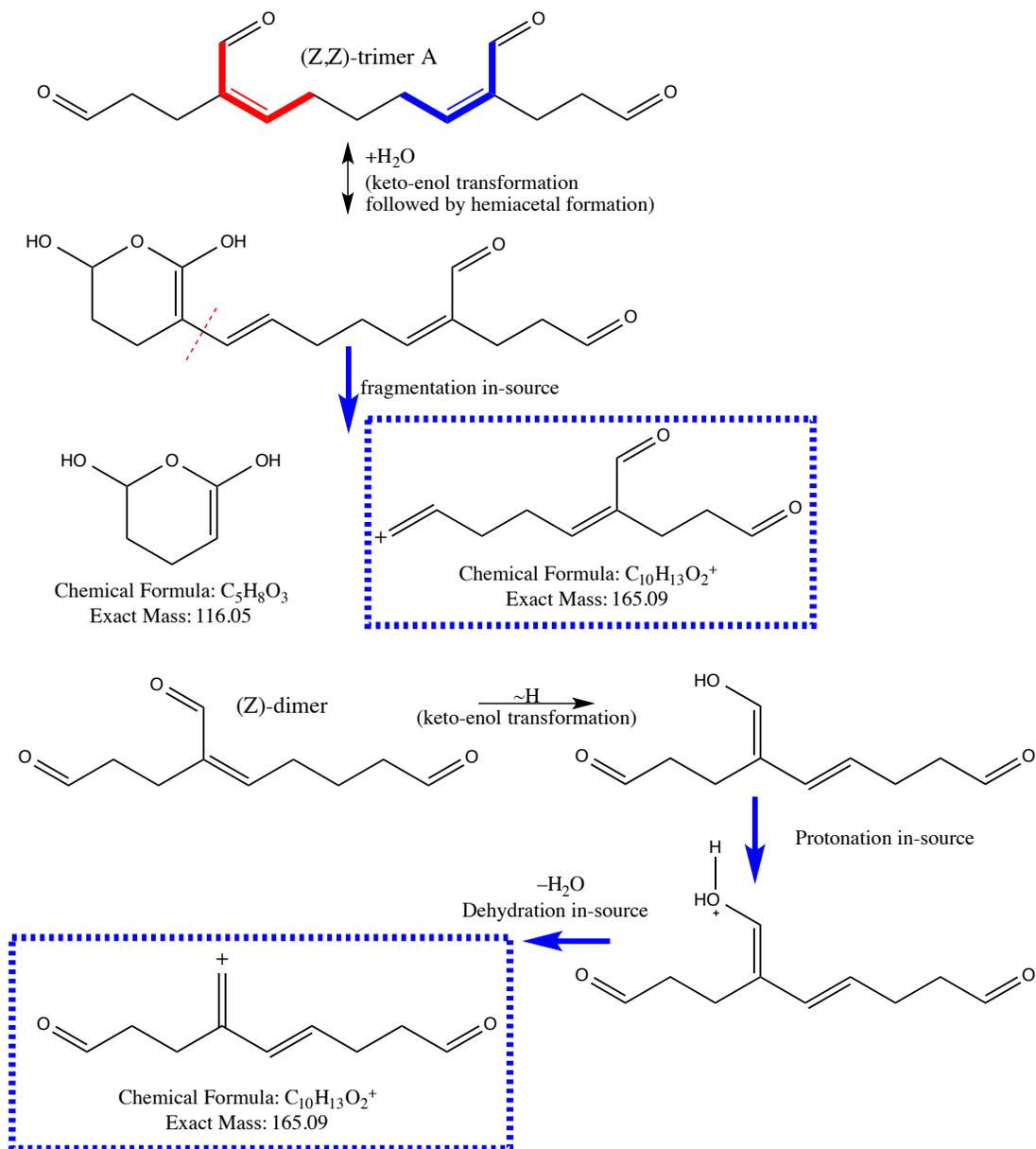


Figure 8. Proposed mass spectrometric fragmentation of GA dimer and trimer.

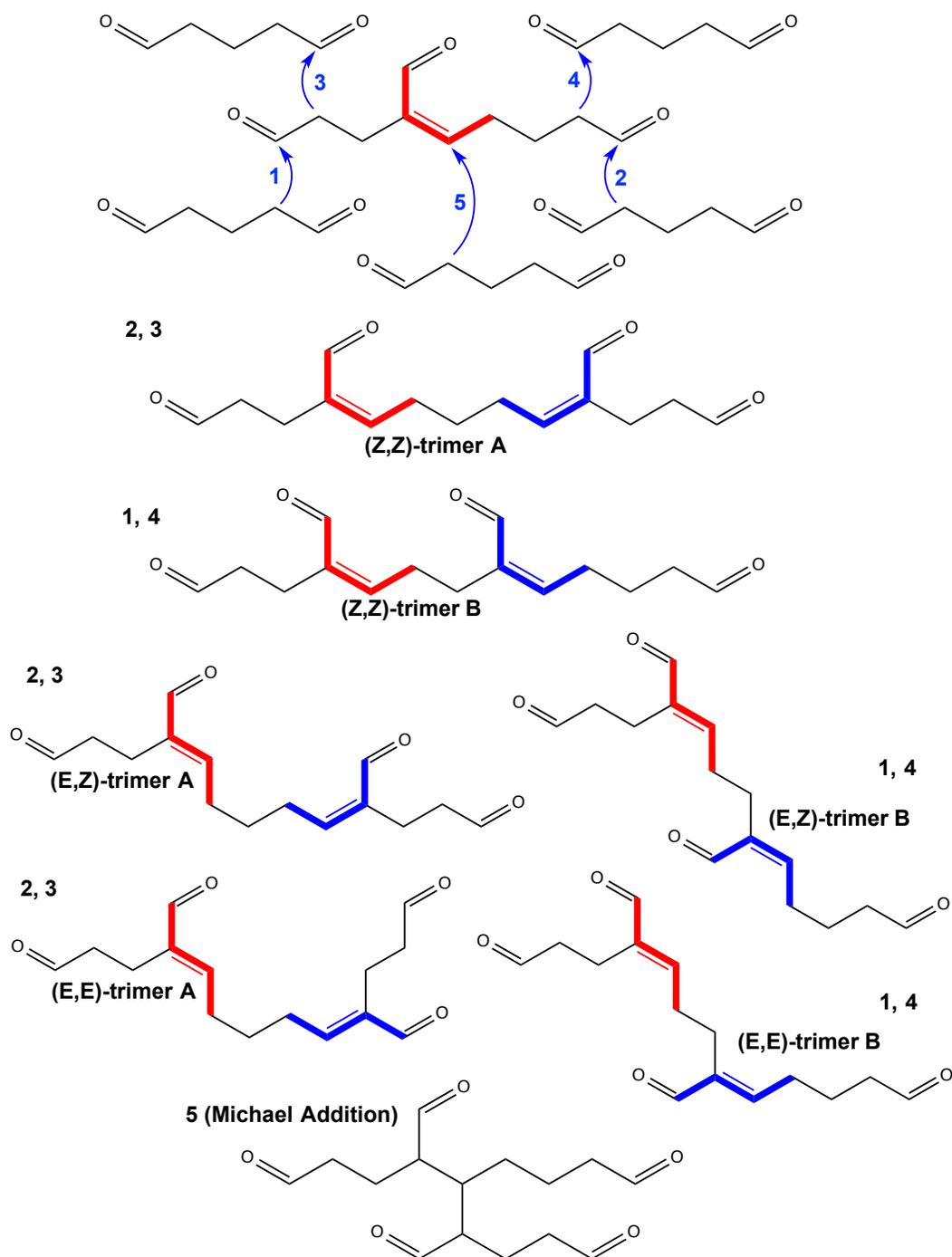


Figure 9. List of the possible reaction sites resulting in the formation of several possible structural isomers of the GA trimer. All molecules shown in full keto form.

Hydration state of GA polymer products. Note that the structures shown in Figure 9 are merely a selection of potential structures possible since there are many possible hydrated forms of each of the dimer/trimer (see Figure 10) and in addition, many different isomers (Figure 9) are consistent with the m/z values obtained. The many peaks in the chromatograms (Figure 11) are suspected to be hydrated/isomeric products; because all keto groups are known to undergo hydration,²⁴⁰ (see Figure 10 for examples) these reactions are suspected to occur downhole. However, because of the nature of electrospray ionization (ESI), the exact level of hydration of polymeric products is extremely difficult to determine, as it is prone to change during the analysis procedure.

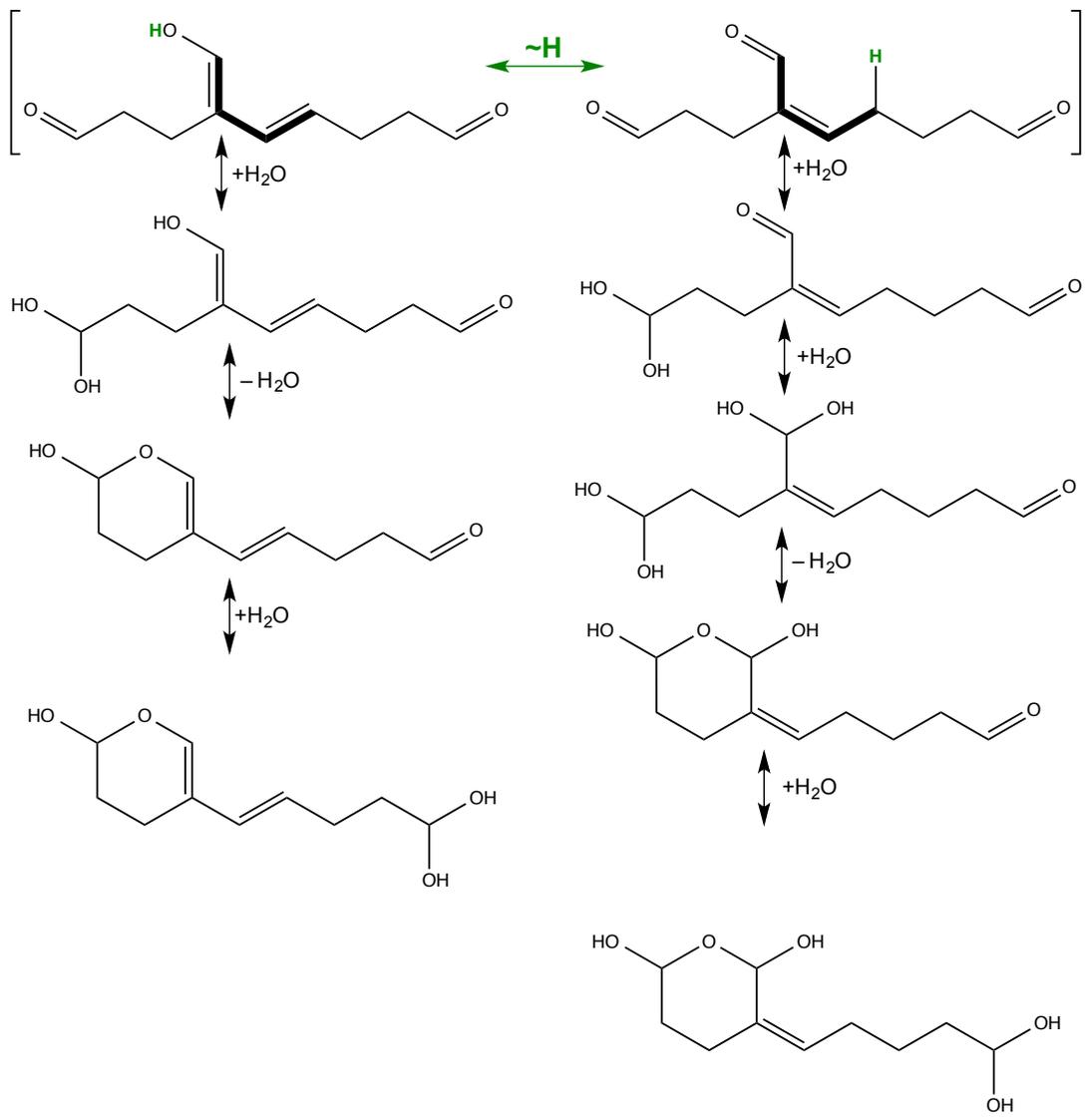


Figure 10. Possible keto-enol, hydration, and cyclic hemiacetal transformations possible for the GA dimer.

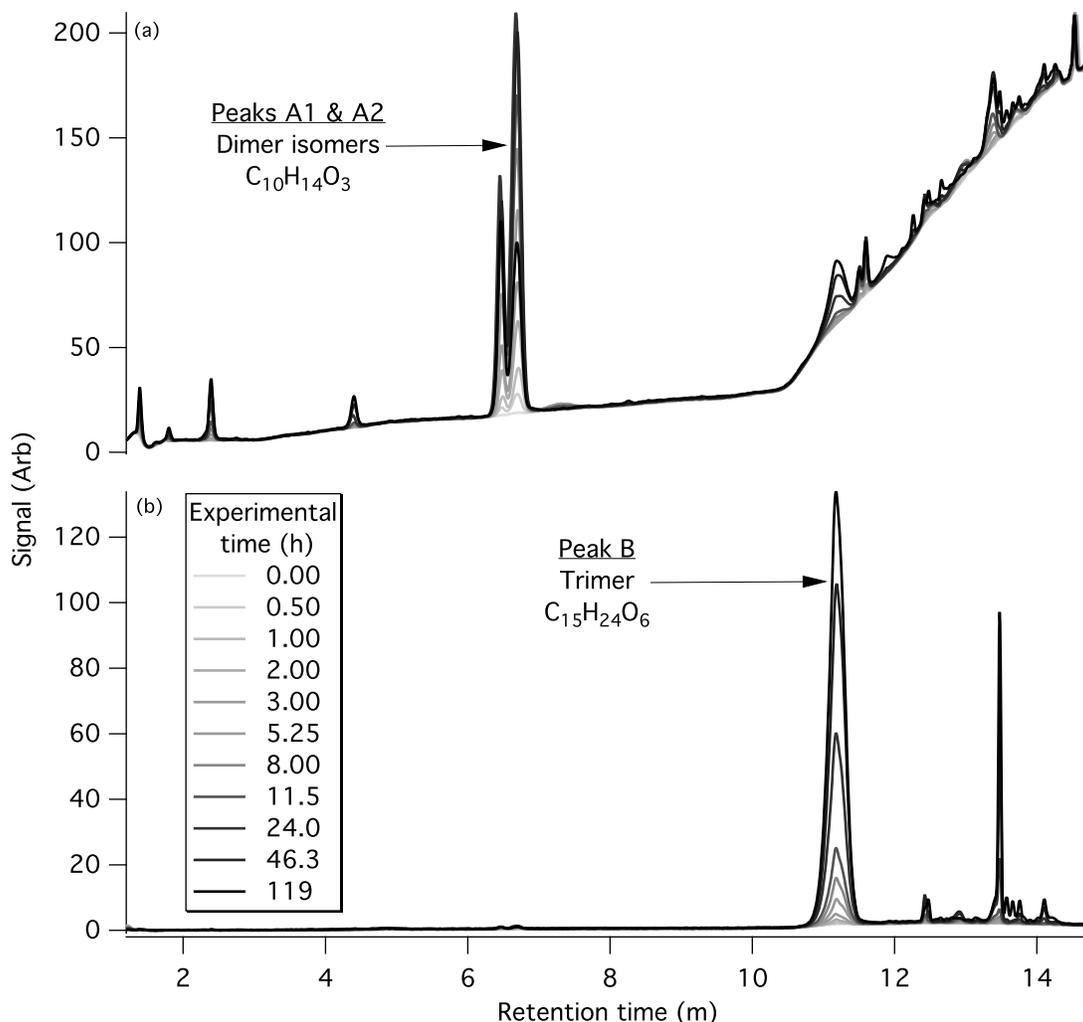


Figure 11. Chromatographic evolution of the transformation products of GA over time.

Kinetic Model. Based on the observation that the matrix parameters affect the chemical kinetics in a systematic and predictable manner, a functional kinetic model based on the Arrhenius equation was generated to approximately predict the transformation of GA as a function of downhole conditions and residence time. The model calculates a pseudo-second-order rate constant k'_{model} ($M^{-1}s^{-1}$) at formation-specific temperature, pH, and salt concentration (as typical major species sodium chloride) via:

$$k'_{\text{model}} = A * e^{-\frac{E_a}{RT}} * [OH^-]^{0.77} * \frac{107.2}{141.8 * F_{\text{NaCl}}[\text{NaCl}] + 107.2}$$

Here, the pre-exponential factor A and activation energy E_a were experimentally determined to be $2.218 * 10^{16} \text{ M}^{-1.77} \text{ s}^{-1}$ and $92,330 \text{ J/mol}$ respectively, R is the gas constant, and T is the temperature (K). The final multiplying factor represents an approximation of the effect of salt on the reaction kinetics where $F_{\text{NaCl}} = 1.2 \text{ M}^{-1}$ is a scaling factor determined via best fit to experimentally observed results. Because of its approximate nature, this salt factor may not necessarily reveal the true nature behind salt's alteration of the chemical kinetics (see below) but should provide adequate as a working best-fit kinetic model for the use of predicting GA degradation under hydraulic fracturing-relevant conditions. The equation for k'_{model} may be simplified into the following unit-less algorithm for use in spreadsheet programs:

$$k'_{\text{model}} = 2.218 * 10^{16} * \exp\left(-\frac{11,106}{T_{\text{c}} + 273.15}\right) * (10^{\text{pH}-14})^{0.77} * \frac{1}{1.5873[\text{NaCl}]_{\text{M}} + 1}$$

where user-input variables have been bolded and include the required units in subscript for ease of use. The full kinetic model then utilizes predicted k'_{model} rate constants in the following second-order relationships to determine half-lives ($t_{1/2 \text{ model}}$) and GA concentrations at any time during downhole residence:

$$t_{\frac{1}{2} \text{ model}} = \frac{1}{k'_{\text{model}}[\text{GA}]_0} \quad \text{and} \quad \frac{1}{[\text{GA}]} = \frac{1}{[\text{GA}]_0} + k'_{\text{model}} t$$

The half-lives predicted (which match experimental half-lives with a correlation of $R^2 = 0.9987$, Figure 12) for the experimental conditions are shown in Table 7. The small error of 3.25% when two parameters are varied from benchmark conditions (here: $140 \text{ }^\circ\text{C}$ and 2 M NaCl) imply that the effects on GA transformation kinetics or rate constant, respectively, are additive.

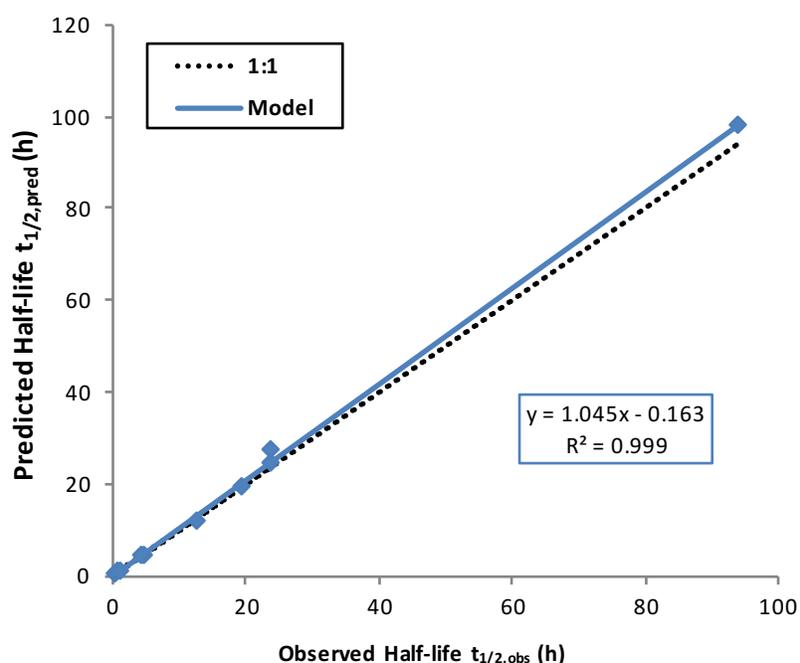


Figure 12. Goodness-of-fit between predicted $t_{1/2}$ values ($t_{1/2, pred}$) on the y axis and experimentally obtained $t_{1/2}$ values ($t_{1/2, obs}$) on the x axis. Also displayed is a 1:1 correlation line for reference.

Effect of Salinity. Far from being simple spectators, salt ions in high concentrations (as measured by ionic strength) are known to alter chemical kinetics in various and often unpredictable ways. These effects are broadly categorized into three general groups: primary, secondary, and ion-pairing salt effects (sometimes called “special” salt effects).^{245,246} Briefly, the primary salt effect describes the changes in chemical reactivity due to the ionic activities in solution, while the secondary salt effect describes how an increasing ionic strength causes an increasing dissociation of weak acids in solution.^{245,246} Ion-pairing effects, on the other hand, describe kinetic alterations produced by interactions between charged salt species and charged reaction mechanism intermediates (such as enolates)— such interactions often impair chemical reactivity and therefore are known to slow chemical kinetics.

In the case of the alteration of GA kinetics, the salt effect is either a secondary or an ion-pairing effect. As the change in chemical kinetics do not follow what would be expected with

changing ionic activities, a primary effect may be ruled out. Both the secondary and the ion-pairing cases, however, are not only explainable by the chemical mechanism (Figure 3) but also matched well with the experimental data.

The secondary salt effect could potentially be inducing the slow-down in kinetics by altering the pH of the base-catalyzed reaction in question. Traditionally, secondary salt effects have been noticed as an acceleration of acid-catalyzed reactions such as hydrolysis.^{245–247} In the case of the base-catalyzed aldol condensation, it is logical to assume that an effect which accelerates an acid-catalyzed reaction will decelerate one which is catalyzed by base; in other words, the secondary salt effect causes less available $[\text{OH}^-]_{\text{aq}}$ with which to catalyze the chemical reaction. Because this step (Figure 7, step 1) is partially rate-controlling as discussed earlier, its impediment would cause the overall kinetics to slow as well. Mathematics of the chemical kinetic equations detailing the secondary salt effect have proven that the relationship between $\log(k'_{\text{obs}})$ and $I^{1/2}$ is linear *only* in the case of the secondary salt effect.²⁴⁷ The data collected here displays an excellent linear fit between experimental $\log(k'_{\text{obs}})$ and $I^{1/2}$ values, with a correlation coefficient of $R^2 = 0.9999$, suggesting that the secondary salt effect is behind the kinetic slow-down observed.

However, ion-pair formation between the enolate and sodium ions may also explain the kinetic slow-down (hence qualifying as an “ion-pair salt effect”). In this scenario, association of Na^+ with the enolate anion decreases its overall nucleophilicity, thereby impeding this rate-controlling step (and therefore k'_{obs}) of the overall chemical mechanism (Figure 7, step 2). Though not often considered a significant effect in polar solvents such as water, ion pairing *is* a recognized phenomenon^{248,249} and ion pairing of enolates is well-documented.^{250–253} The reason this effect may come into play despite the polar solvent is due to the unique nature of highly

saline solutions— due to salt’s drastic depressive effect on electrical permittivity (ϵ) of water ($\epsilon \approx 50$ at 3 M NaCl),²⁵⁴ a super-saline solution may act more like a non-polar organic solvent than non-saline water, thus increasing the likelihood of an ion pairing effect. The data also display a good fit to support this theory, showing a linear correlation between k' and $1/[\text{NaCl}]$ (the mathematically proven relationship which is linear when ion-pairing salt effects are at work^{245,246}) with a coefficient of $R^2 = 0.9967$; while not as strong a correlation as displayed by the secondary salt effect relationship, it is strong enough that the option of an ion-pairing salt effect cannot be ruled out. Only more quantitative analyses and further collection of data for precise chemical mechanism(s) will reveal the true nature of this salt effect, and whether or not it is already well-known in chemistry.

Environmental Implications. The demonstrated effect of the deep subsurface matrix parameters temperature, pH value, and salinity on the transformation of the biocide GA implies that the assessment of any potential environmental impact upon inadvertent release of GA-containing HFF depends on whether a spill occurs before or after HFF injection and on the nature of the injection itself (Figure 13). Pre-injection, only the unreacted GA monomer has to be targeted (for instance by deactivation with bisulfite).⁷² In case GA leaches into groundwater or contaminates surface water, it may be diluted or react with amine or thiol functional groups in natural organic matter until its concentration has diminished past the point of being toxic (LC_{50} over 48h is 0.35 mg/L for *Daphnia magna*).²⁵⁵ When below an inhibitory concentration, GA may readily biodegrade into non-toxic byproducts.^{22,256}

However, once GA is injected underground, it is subjected to the unique deep subsurface environment for several days, during which it may not only react with microbes and both dissolved and solid-phase nucleophiles, but will also react with with another GA molecule

(Figure 4) as dictated by the geophysical parameters temperature, pH, and salinity. Barring significant cooling of the underground environment due to mixing with influent water (Figure 4), the research presented here indicates that GA most likely undergoes rapid autopolymerization (and/or reaction) when used in hot ($T \geq 100$ °C), relatively non-saline shales such as the Haynesville in Texas ($T_{\text{avg}} = 157$ °C). Here, the half-lives of the aqueous GA monomer are on the order of a few hours or less due to autopolymerization alone. Based on the gathered observations, the larger produced polymers will likely sorb to the shale and remain underground instead of resurfacing with the flowback water. Nevertheless, smaller GA oligomers may still resurface as observed in flowback and produced water samples from Colorado.²³⁶ Downhole temperatures in the Wattenberg Field in CO have been reported to exceed 115°C ²⁵⁷ and the majority of wells has <35 g/L TDS (Figure 4). Unfortunately, the GA dimer's and trimers' potential environmental impact and toxicity is largely unknown. Previous research indicates that the oligomeric products of GA do retain some protein-crosslinking capability^{145,258}, but they are likely to have lessened reactivity as compared to the monomer based on steric considerations and the fact that one (active) aldehyde group is consumed in each polymerization step.

In milder ($T \leq 60^{\circ}\text{C}$), neutral to acidic, and/or saline shales (such as the Marcellus), autopolymerization is not expected to play a large role in GA's transformation, and aqueous removal of GA will most likely be dominated by reactions with target bacteria and possibly other HF fluid additives.²² Nevertheless, in absence of rapid autopolymerization, the biocide will likely remain stable for a substantially longer period of time. For instance, at 65 °C, neutral pH, and a concentration of 2 M NaCl (i.e., in a typical range for the Marcellus Shale), the model predicts a half-life of 20 days at an initial GA concentration of 500 mg/L. This implies that under these conditions the returning flowback water may still hold substantial, actively biocidal

GA concentrations as well as some aqueous GA oligomers that may impair subsequent water treatment processes or natural attenuation processes upon accidental releases.

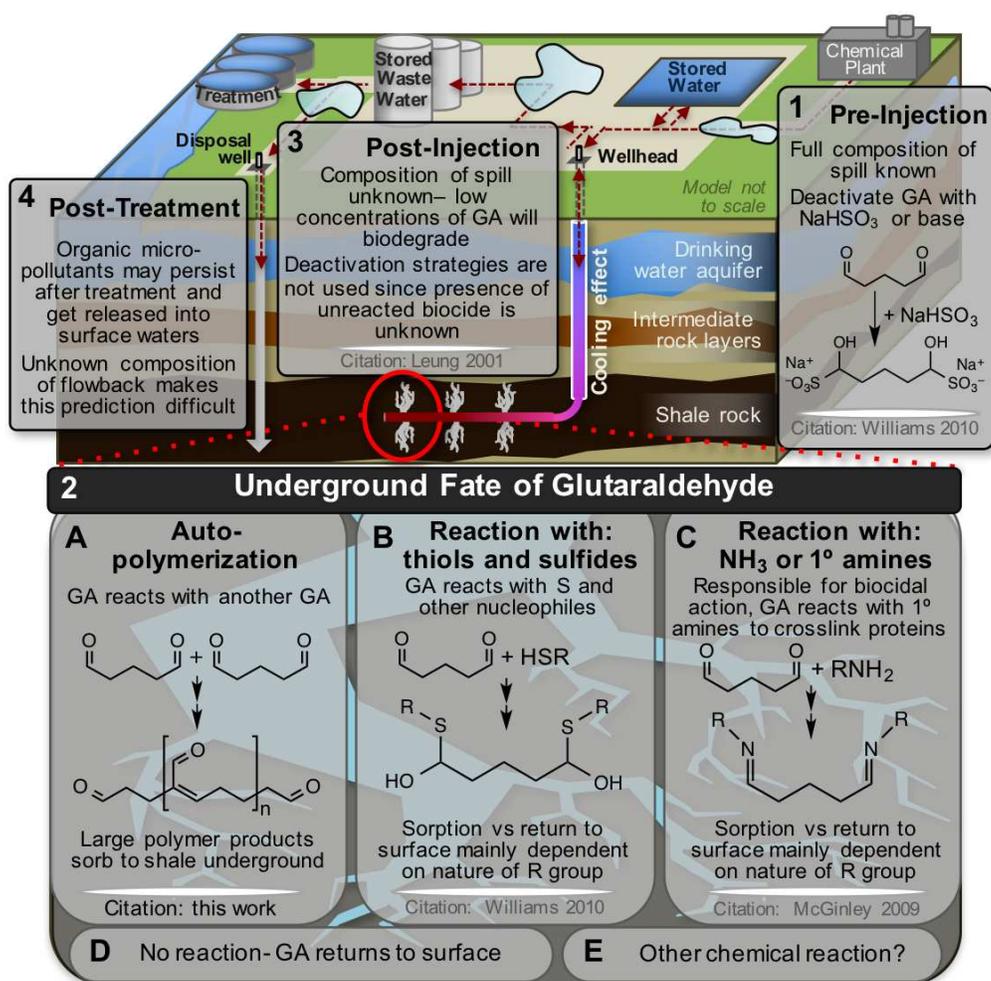


Figure 13. Environmental fate and transport of GA with regards to its use in hydraulic fracturing, with special emphasis on downhole fate. Citations, when provided, represent research that greatly contributed to the knowledge referenced and is not necessarily all-inclusive. Also displayed is the cooling effect caused by injection of cold fracturing fluid at the surface level.

Our findings highlight the fundamental role of deep subsurface chemistry in controlling the downhole fate of organic HFF additives. The transformations of the biocide glutaraldehyde (GA) as a model compound have been explored, but depending on the chemical composition,

structure, and speciation of other additives, several other mechanisms may produce reaction intermediates that are unique to these extreme underground environments. For instance, water-driven redox reactions, (de)hydrations, and (de)hydrogenations have been reported to occur in hydrothermal vents at temperatures over 300 °C and pressures around 100,000 kPa.^{175,176,255} Additionally, parameters such as the dielectric constant (ϵ) change drastically in extreme conditions; for instance, increasing the temperature of water from 25 to 300 °C causes ϵ to fall from 80 to 20 (roughly the ϵ of acetone).¹⁷⁷ Decreasing ϵ to such an extent may significantly impact reactant solubility and thus result in different chemical reactions. Lowering ϵ to this degree also has a substantial impact on reaction kinetics since ion pairing effects are greatly exacerbated, as is explored above with the salt effect.²⁵⁶ Previous research has shown that divalent cations in a supercritical environment hold anions in ion pairs very closely, just beyond the first hydration shell at 425 °C,²⁵⁷ and at many points past supercritical ($T > 374$ °C, $P > 22,060$ kPa), water basically behaves as a non-polar solvent.²⁵⁶ Conditions that are both hot *and* saline could potentially have a considerable impact on downhole organic chemistry, as both factors serve to lower ϵ . However, since most current research focuses on inorganic components alone, very little is known about the effects of ion pairing on organic compounds and reactions at such extreme conditions.

Incidentally, the chemical reactions that uniquely run only in high temperatures and pressures due to the chemi-physical alterations of the solvent quickly cease under milder conditions (<250 °C),^{176,177} because it is the unique physical changes the *solvent* (in this case water) undergoes as a result of the extreme temperature/pressure which enable these unique chemical reactions to occur (instead of merely provision of activation energy), these reactions cannot occur at more mild conditions with “normal” solvent behavior. Therefore, within

conditions relevant to hydraulic fracturing, it is likely that temperature and pressure only affect reaction kinetics and *not* mechanisms and pathways (perhaps with the exception of very hot and pressurized fracturing operations in extremely deep shales such as the Haynesville in Texas). Salinity, on the other hand, is expected to play a major part in all of the downhole chemical reactions of HFF additives in all shales, shallow or deep. For instance, hydrolysis is one of the most common mechanisms by which molecules are transformed in aqueous surface environments. However, hydrolysis reaction kinetics *and* pathways may be altered at high salinities. It is well established that at high concentrations, chloride and bromide may compete with water in nucleophilic substitution reactions,²⁵⁸ potentially producing organohalogen compounds during reaction with susceptible organic HFF additives downhole. Because many organohalogens are toxic even at low concentrations, their potential presence within produced water may pose risks to the inhabited surface environment.

Clearly, more research is critically needed to obtain a better understanding of deep subsurface transformation of other HFF additives at the extreme temperatures, pressures, and especially salinities in unconventional shale reservoirs. Even ten years after the exemption of underground HFF injections from the Safe Drinking Water Act (SDWA), hardly anything is known about the variety of organic chemicals resurfacing in flowback water, and current regulation - if it exists at all - only targets naturally occurring, formation-derived species such as petroleum hydrocarbons. Without a doubt, the multifaceted deep subsurface conditions present in hydraulic fracturing operations play a key role in the accurate prediction of the chemical efficacy and environmental fate of HFF additives. Hopefully this first-of-its-kind investigation and predictive model development will spark new initiatives on developing more sustainable

HFF additives and researching their downhole fate *before* they are introduced into the environment.

CHAPTER 4 – DOWNHOLE TRANSFORMATION OF THE SURFACTANT NONYLPHENOL ETHOXYLATE

INTRODUCTION

Nonylphenol ethoxylates (NPEs) are a class of toxic chemicals commonly used as surfactants and corrosion inhibitors in oilfield operations. As many as 50% percent of all currently existing wells report using NPEs within their hydraulic fracturing fluid (HFF) formulations.⁶ Despite NPEs' effective and very powerful surfactant and corrosion inhibition properties, growing suspicion over their possible release of nonylphenol (NP), a known endocrine disrupting compound (EDC), clouds its current use in industry.⁶ NPEs and NP are known environmental contaminants^{259–262} and have been widely observed in aquatic and terrestrial environments including wastewater treatment plants,^{260,263–265} natural river waters and sediments,^{259,263,265–267} living fish tissue,^{259,268,269} and agricultural soils treated with sewage sludge.^{270,271} NP is one of the most commonly found pollutants in air and dust within indoor environments.²⁶⁰

The aerobic biodegradation of NPEs into NP is well studied and indicates that microbes slowly (half-lives of months to years) transform NPEs into NP by successive cleavage of terminal ethoxylate subunits.^{259,266,272–275} However, in the absence of oxygen, the biodegradation of NPEs and other ethoxylated alkylphenols is far less studied, and results are varied. Anaerobic biodegradation appears to occur extremely slowly, with half-lives in the order of years, increasing based on the PEG chain length.^{260,276} Other research suggests that steric factors of highly branched C₉ groups in NPEs may further hinder anaerobic biodegradation,²⁷⁷ though such results have not been verified.

The fate of NPEs under abiotic conditions, however, is less understood. The few studies reporting on NPE degradation in abiotic surface environments show that NPE is unreactive, and likewise does not degrade during short (2-hour) thermal degradation-hydrolysis attempts.^{260,276,278,279} The behavior of NPE under the unique conditions in unconventional reservoirs, however, has not been examined yet. Downhole, HFFs become heated, pressurized, saline to varying degrees, and (ideally) abiotic and anoxic due to the addition of biocides and oxygen scavengers^{28,280} HFFs are then held under these conditions for several days, and possibly for even longer periods of time if the petroleum recovery process is paused due to unfavorable economics.

The downhole temperature that HFF additives are subjected to for this extended time period ranges drastically from shale to shale from 40 °C in parts of the Marcellus to 193 °C in parts of the Haynesville.^{280,281} Total dissolved solids (TDS) are often observed at high levels within flowback water but vary drastically between shales; comprised mainly of dissolved Na⁺, Ca²⁺, and Cl⁻, the TDS levels of produced water from more saline shales such as the Marcellus and Bakken can exceed 300,000 mg/L.^{8,15,24}

In light of this vast knowledge gap preventing any environmental and/or human health risk assessment with regards to the use of NPEs in hydraulic fracturing, the aim of this study was to reveal the fate of NPEs under simulated oil & gas reservoir conditions in high pressure and temperature stainless steel reactors. The temperature chosen for this study (100 °C) represents the low-end cutoff for a “hot” shale,¹⁸ and has also been shown to be too high to sustain bacterial activity downhole,²⁸⁰ ensuring an abiotic environment. The results gathered here are consistent with field studies^{13,282} that have revealed the presence of not only PEGs²⁸² in flowback water, but also NP and other alkylated phenols at concentrations up to 7.9 µg/L.¹³ Thus, our findings also

serve as an important fundamental starting block in the quest for a better understanding of flowback and produced water quality and their potential adverse environmental and health effects.

EXPERIMENTAL

Reaction Solutions. Stainless steel reaction vessels (model no. 4768, Parr Instrument Company, MAWP 3,000 psi/200 bar at 350 °C) were used to hold various NPE solutions at simulated downhole conditions (see Figure 14 for details) while samples were collected over time. Detection and analysis described in SI. All chemicals were of ACS grade (Sigma-Aldrich, St. Louis, MO) unless otherwise stated. 4-nonylphenol standard was purchased from Supelco (Bellefonte, PA). NPE solutions were prepared in 18 MΩ DI H₂O (Millipore, Billerica, Massachusetts) at a concentration of approximately 0.15 mM, matching a working 100 mg/L concentration. pH was controlled at pH = 5 via 25 mM phosphate buffer. Synthetic brine (0.5 M NaCl) and shale samples (average surface area 15.0 m²/g, 9.1% organic content) described in more detail in previous work.²⁸⁶

Electrospray Ionization Time-of-Flight Mass Spectrometry (ESI-TOF-MS). Mass spectra were recorded with an HPLC (1200 model, Agilent Technologies, Inc.) coupled to a time-of-flight mass spectrometer (TOF-MS) (6220 model, Agilent Technologies, Inc.). The samples were separated using a C18PFP 2.1x150 mm with 3.0 μm particle size column (Phenomenex, Torrance, CA) prior to TOF-MS detection. HPLC conditions were as follows: Injection volume used was 30 μL, flow rate was 0.5 mL/min using solvents A- water plus 0.1% formic acid- and B- acetonitrile. Gradient elution was as follows: 5% B from 0 to 2 minutes, 5% to 25% B from 5 to 25 minutes, 25% to 95% B from 5 to 13 minutes, then hold 95% B until 20 minutes, after which the column is re-equilibrated to 5% B for another 5 minutes, bringing total runtime to 25

minutes. The instrumental conditions for the ToF-MS were: capillary voltage 4,500 V, skimmer voltage 60.0 V, fragmentor voltage 100V. Cone gas was N₂, held at 310 °C, 8.0 L/min, with a nebulizer pressure of 310 kPa (45 psi). Data were collected between a mass range of 90-2,000 with a scan rate of 1.05. All mass spectra collected in negative ion mode.

Nuclear Magnetic Resonance Spectroscopy. The NMR study was done on a Varian Inova 600 MHz ¹H NMR equipped with four channels and a sensitive 5 mm HCN probe. Sample was dissolved in 550 µl of CDCl₃ and placed in a Wilmad 5mm-535pp NMR tube. Probe temperature was held at 25°C, aided by an FTS Chiller providing -5 °C conditioned N₂ gas to the probe head. The Varian standard S2PUL pulse sequence was used in a simple one pulse and acquire mode. No sample spinning was used. The spectral window used was 9600 Hz, 16 K complex points were acquired with an acquisition time of 1.7 s followed by a delay of 2 s for an overall 3.7 s recycle time between transients. A $\pi/2$ pulse of 9.6 µs was used to excite the protons. Two steady-state pulses were done followed by eight signal averaging transients for each sample. The embedded digital signal processor was on and used to correct the raw, time-domain data in real time. The data was Fourier transformed without zero-filling or any smoothing applied. Spectral referencing of the frequency domain data was done by the software using the value of the CHCl₃ impurity in the spectrum as 7.25ppm.

RESULTS AND DISCUSSION

As shown in Figure DEG, when subjected to a hydraulic fracturing-like environment— anoxic, 100 °C, pH 5, and pressurized at 20 bar— the hydraulic fracturing chemical NPE hydrolyzes at the oxygen proximal to the benzyl ring, creating a PEG chain and the endocrine-disrupting compound NP. Even though this breakdown has been previously predicted,⁶ other

studies have failed to demonstrate the hydrolysis of NPE into NP: the reason for this discrepancy is only due to the time the chemical is subjected to extreme heat and pressure. In no environment outside of hydraulic fracturing is NPE held at continual high heat, pressure, and acidity for days to weeks at a time; therefore, this set of conditions has never been tested before, as there was no need.

The results presented here reveal for the first time that in anoxic environments at 100 °C, hydrolysis is the major transformation mechanism by which NPEs degrade into NP (Figure 15). Chemical degradation was abiotic, given the extreme temperature and pressure; to ensure no microorganisms survived the autoclave-like conditions within the vessels, a test for ATP verified it was below limits of detection (data not shown). The mass spectrometry results of the NPEs and their transformation products are shown in Table 8, and their temporal changes are illustrated in Figures 14 and 16. The high degree of branching in the C₉ chain of the NPE starting material (Figure 17) did not inhibit transformation of NPEs into NP as has been suggested for anaerobic biodegradation.²⁷⁷

Hydrolysis of ethers in general is a kinetically slow process ($t_{1/2}$ from weeks to months²⁵⁸) unless the temperature is drastically elevated, as is the case downhole; wells drilled in 10,000-foot deep shales such as the Haynesville in Texas can reach temperatures of 200 °C.²⁸ In the reactor simulations run without salt or shale at 100 °C and pH 5, NP was detected in low levels in all NPE solutions (even within the starting solution) and continually increased over time in the heated and pressurized environment while the concentration of NPEs steadily decreased (Figure 14). The presence of NP contamination in the original commercial NPE source is itself a significant finding, something which has been previously reported of commercial-grade NPE solutions.²⁷⁵ Over time during the downhole simulation, all polymer sizes of NPEs appeared to

decrease proportionally to each other, indicating that terminal PEG subunits are *not* being successively cleaved as is the case in NPE biodegradation.^{259,266,272–275} Also considering that PEGs were detected at polymer lengths exactly consistent with the hydrolysis of NPEs at the first oxygen proximal to the aromatic ring (Figure 18), it can be deduced that NPEs hydrolyze directly into NP and a PEG the length of the original polymer tail (Figure 15). Similarly, in studies of other PEG derivatives, it has been shown that with addition of heat the PEG chain separates from the molecule via cleavage at the oxygen exactly proximal to the derivatized leaving group²⁸⁴ further lending strength to the analogous mechanism proposed above (Figure 15). It is also important to note that it can be expected that other strong nucleophiles (such as primary amines) will likewise react with protonated NPEs, similarly producing NP and a PEG substituted with the given nucleophile.²⁴⁰

That the chemical reaction is initiated at the oxygen proximal to the benzyl ring (or in the “⟨” position”) is consistent with the electronic environment of the NPE molecule. The benzyl ring is highly electronegative and the oxygen is connected to this system; essentially its lone-pair oxygens overlap with the pi-conjugation system of the benzyl ring (it’s an electron-donating substituent). This makes the ⟨-oxygen especially partially-negative (especially when compared to the other oxygens in the PEG chain), which not only facilitates nucleophilic attack on a hydronium (Figure 15) but also lends stability to the subsequent transition state, thus lowering the activation energy needed for chemical reaction at the ⟨-oxygen only.

The presence of salt in the form of NaCl had a dramatic effect on the degradation of NPEs. According to the secondary salt effect,²⁴¹ it is expected that an increase in salt concentration will effectively lower the pH in solution resulting in an increased rate of (acid-catalyzed) hydrolysis; this salt phenomenon has previously been shown to have significant

effects on hydrolysis reaction rates of other organic compounds.^{285,286} An effectively lowered pH thereby implies that increased salinity would result in an increased production of NP within a shorter time period, which is indeed the case observed within the simulation vessels (Figure 19). However, presence of a “special salt effect” whereby the ion-pairing of Cl^- and protonated intermediates^{248,249,250–253} cannot be ruled out given the data gathered here; further research is required to elucidate the full nature of the salt effect observed here.

This is not the first time that salinity has shown to have a significant effect on the degradation of a fracturing fluid additive within a simulated downhole environment; it was previously discovered that NaCl concentration stagnates the auto-condensation of glutaraldehyde,²⁸³ not surprising as that process is base-catalyzed. The acceleration of NP generation observed due to NaCl has significant environmental implications due to the fact that the downhole environment can be extremely saline depending on the shale in question,²⁸ often far more saline than the 0.5M NaCl condition tested here, meaning NP will be produced faster in fracturing operations located in ultra-saline shales (such as the Marcellus and Bakken). However, an aqueous solution which is increasingly saline is also similarly less hospitable to nonpolar species, meaning that even though NP may be produced at a faster rate, it will be less soluble, especially in the presence of sorbents such as shale.

As has been previously alluded, the main control on the aqueous NP concentration is sorption effects.²⁶⁰ This seems to be similarly true of NPEs as their concentration quickly drops to below the limit of detection of the method used (\oplus 0.05 ppm) in simulations containing shale (Figure 20). Notice that in the presence of salt (Figure 20), the NPE concentration drops dramatically- more than can be explained by transformation to NP- and then stays fairly constant, suggesting a major “salting out” sorption effect even without shale, and the presence of

a better sorptive medium (shale) only enhances this effect. Because the ether linkages in NPE are still expected to be in contact with solvent water molecules when adsorbed to a solid surface, it is likely that NPEs will still undergo hydrolysis much as is seen in the absence of shale (while full absorption into shale natural organic matter—thus escaping water—is technically possible, it is not likely given the polar PEG tail of the NPE molecule²⁵⁸). Whether the degradants desorb from the shale, however, is a completely different story. In the time frame of the experiments performed for this study (approximately 1,000 hours), no desorption of NPEs, NP, or PEGs were observed in the simulator (Figure 20). Indeed in other environments such as groundwater it has been noted that due to its low solubility and high tendency for sorption, NP has a far lower rate of mass transfer than other, more soluble pollutants, and that sorption of NP is controlled by the percent of organic content in local sediments.²⁶⁰ If either degradant were to desorb, it would most likely be the oxygen-rich PEG chain, which would match real-world observations of PEG species returning with flowback wastewater.²⁸²

Table 8. Major chemicals identified at various time points throughout experimental runs, including both starting materials (NPEs) and degradation products (NP and PEGs). Note that NPE and PEG chain length is designated by its ‘n’ number.

Retention Time (min)	Measured Mass (<i>m/z</i>)	Ion Formula	Base Peak	Putative Identification	Theoretical <i>m/z</i>	Error (ppm)
12.09	219.1755	C ₁₅ H ₂₃ O ⁻	[M-H] ⁻	Nonylphenol	220.1828	-0.25
11.89	793.5107	C ₄₄ H ₇₃ O ₁₂ ⁻	[M+COOH] ⁻	Nonylphenol Ethoxylate n=12	748.5126	0.57
11.89	749.4843	C ₄₂ H ₆₉ O ₁₁ ⁻	[M+COOH] ⁻	Nonylphenol Ethoxylate n=11	704.4918	0.45
11.89	705.4570	C ₄₀ H ₆₅ O ₁₀ ⁻	[M+COOH] ⁻	Nonylphenol Ethoxylate n=10	660.4649	-1.07
7.53	591.3386	C ₂₉ H ₅₁ O ₁₂ ⁻	[M+COOH] ⁻	Polyethylene Glycol n=11	546.3404	-8.73
7.35	547.3124	C ₂₇ H ₄₇ O ₁₁ ⁻	[M+COOH] ⁻	Polyethylene Glycol n=10	502.3142	-4.86
7.14	503.2862	C ₂₅ H ₄₃ O ₁₀ ⁻	[M+COOH] ⁻	Polyethylene Glycol n=9	458.2895	-3.38

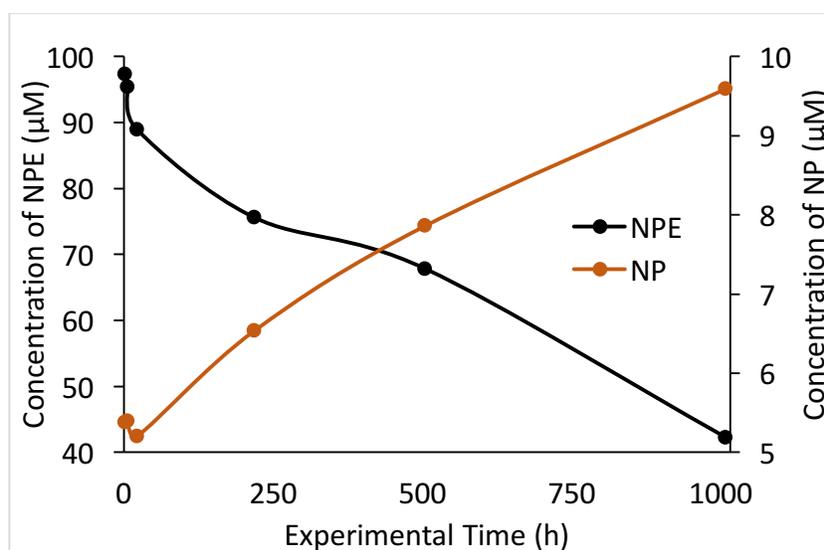


Figure 14. Degradation of NPE with concurrent generation of NP over time. Reaction vessels were maintained at 100°C, pH 5, and 20 bar without salt or shale. Note that only the 3 most significant polymers of NPE (listed in Table 8) were used for quantification.

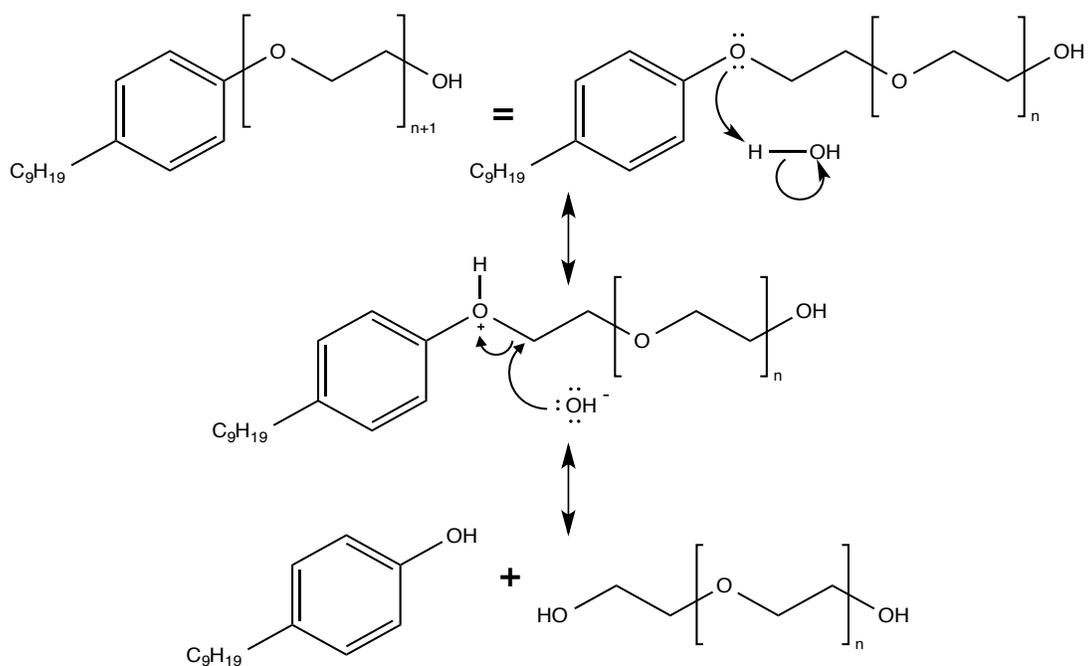


Figure 15. Proposed mechanism of NPE transformation into NP under downhole conditions; acid-catalyzed hydrolysis localized on the oxygen proximal to the aromatic ring. Note that the difference in “n” values between polymer chain lengths of NPE and PEGs (as seen in Table 1) is merely due to differences in nomenclature between the two species; the degradant PEG chain remains the same length as it was originally in NPE despite its smaller “n” value, which here has been fully drawn out for clarity.

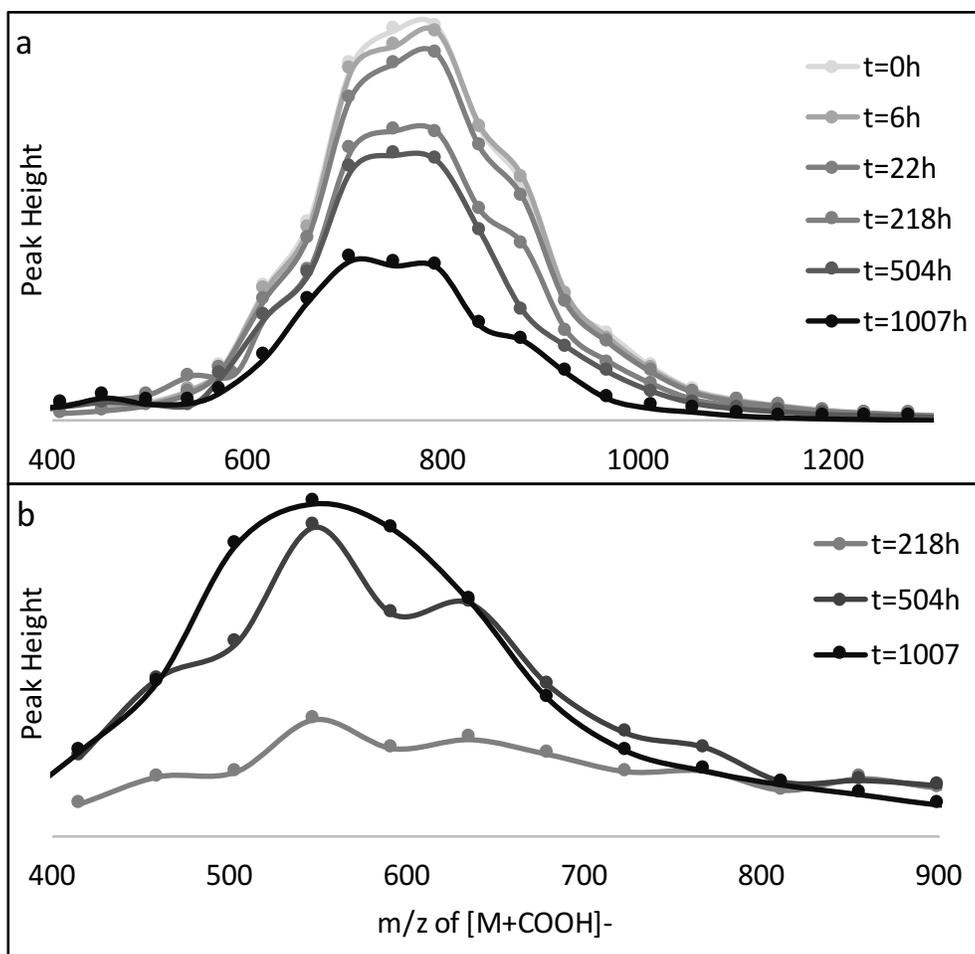


Figure 16. (a) Degradation of NPEs and (b) generation of corresponding PEGs over the experimental time. While the line coloration indicates experimental time, the x axis indicates m/z of the molecule. It can be seen from each graph that all sizes of polymers degrade and generate evenly, suggesting that PEGs hydrolyze cleanly from NPE at the oxygen most proximal to the aromatic ring.

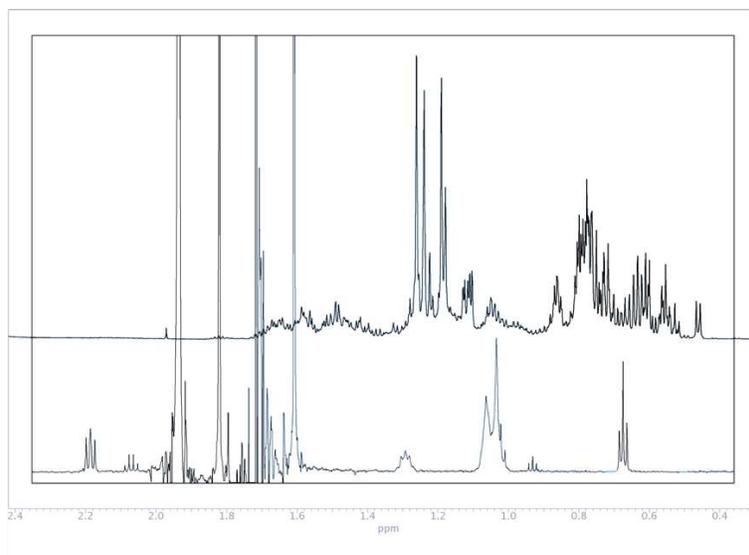


Figure 17. Displayed above are portions of the NMR spectra of the NPE starting material (top) and the 4-NP standard (bottom). Notice the complexity of the NPE starting material below 1.0 ppm as compared to 4-NP; this part of the NMR spectrum is only populated by terminal methyl protons. The simplicity of the bottom spectrum is indicative of the fact that the 4-NP standard has a verified linear C9 group with only one type of terminal methyl proton, whereas the complexity of the top spectrum indicates that the NPE's C9

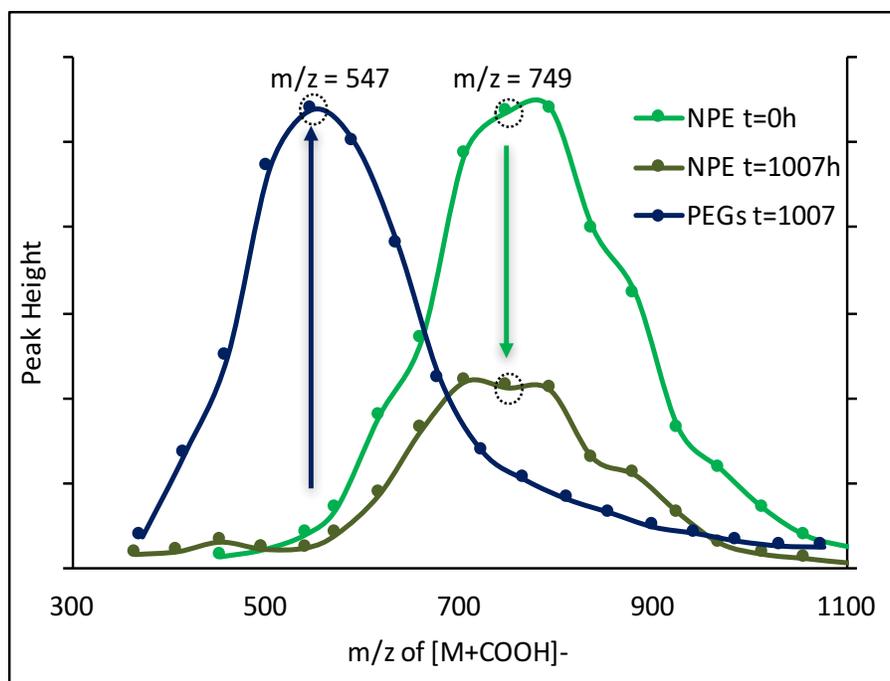


Figure 18. Degradation of NPE (green lines) vs. generation of PEGs (blue line) at 100 °C, pH 5, in the absence of shale and salt. Notice the mass difference between the primary NPE polymer degraded (m/z 749) and the primary PEG generated (m/z 547) plus one water molecule (18 amu) is exactly the mass of NP (220 amu). This indicates complete conversion of NPE into NP via hydrolysis.

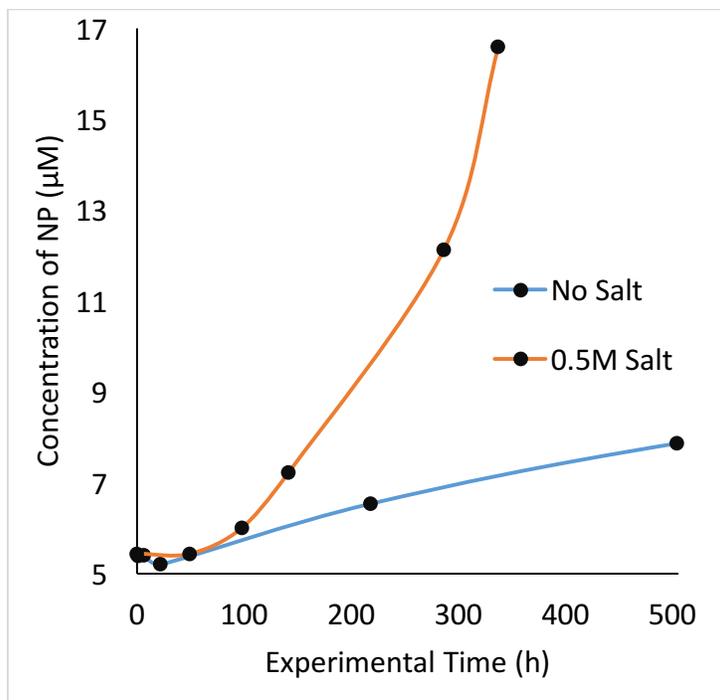


Figure 19. Displayed above is the effect of addition of the addition of NaCl to the simulator vessels on generation of NP, shown in μM (y axis) over time in hours (x axis). Notice the drastic acceleration of NP generation, a result of salt's effective lowering of the pH on an acid-catalyzed reaction, a phenomenon called the secondary salt effect.

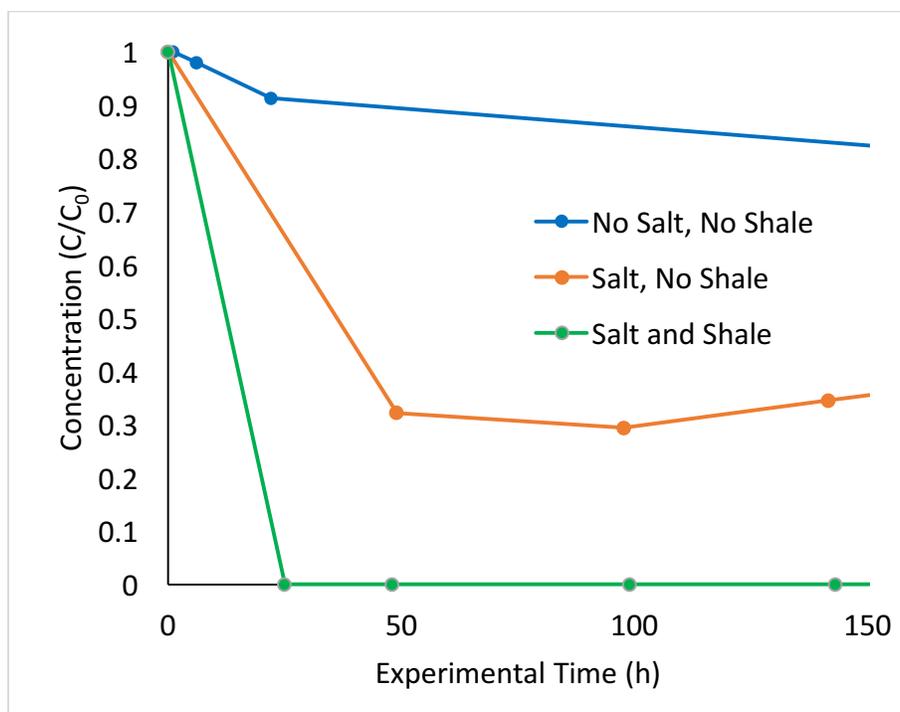


Figure 20. Shown above is a comparison of the effects of salt (0.5M NaCl) and presence of shale on the removal of NPE from aqueous solution at 100°C and pH 5. Removal from solution is controlled both by chemical degradation and by sorption; clearly, sorption has a strong effect when shale is introduced into the simulators.

Environmental Implications

It is shown here for the first time that under simulated downhole conditions relevant to hydraulic fracturing, NPEs are hydrolyzing directly into PEGs and NP, something only recently predicted as possible in another critical review by Elsner et. al.⁶ The observed high sorption affinity of the problematic NP to organic-rich mineral surfaces such as shale imply that the majority of NP will sorb underground and remain effectively removed from the water column during hydraulic fracturing. However, even small concentrations of EDC's are cause for concern, and therefore even a small amount of leaching is significant. In fact, as mentioned earlier, the presence of alkylated phenols including NP have been detected in produced waters from hydraulic fracturing at concentrations up to 7.9 µg/L.¹³ While these authors suggest that

NP may be a product of biodegradation, it is also entirely consistent with the mechanism proposed above (Figure 15).

In fact, the exceptional sorption of NPE to shale may ensure that the molecule stays underground and under heat long enough to degrade into NP, when otherwise a hydraulic fracturing event might end in a shorter timescale than is of concern for this hydrolysis reaction. The observation of PEGs in produced water²⁸² certainly suggest this is possible, since these water-soluble substances should wash up immediately with flowback water; the fact that they are being detected over a period of time and that the PEGs detected are chemically different than any which were initially injected is also highly suggestive of a breakdown process, such as the one we have suggested above (Figure 15). Assuredly, further research is needed to assess this potentially serious breakdown process to determine the role it will play in disposal and treatment of wastewater from oil and gas recovery operations.

CHAPTER 5 – CONCLUSIONS AND FUTURE WORK

The overarching goal of this dissertation was to examine the downhole behavior of selected HFF additives as well as providing the framework for future experiments on other chemicals. The selected HFF additives, GA (glutaraldehyde) and NPE (nonylphenol ethoxylate) are both widely used in hydraulic fracturing operations. Not only are they exposed to a huge range of downhole conditions, but the large quantities being used means a large chance of transformation products potentially ending up in hydraulic fracturing wastewater, or flowback.

Chapter three not only laid out the framework for laboratory testing of downhole matrix parameters, but was the first research actively searching for transformation products of the organic additives used in hydraulic fracturing. This research demonstrated that, even in the absence of viable nucleophiles, GA will undergo aldol condensation with itself to create long, insoluble polymer chains. Because environmental research prior to this theorized that GA underwent degradation via hydrolysis,²⁵² the confirmation that the chemical instead undergoes aldol auto-condensation is significant especially from a kinetics standpoint; whereas hydrolysis proceeds via first-order kinetics, this research clearly reveals that GA degrades following second-order kinetics, which is significant when attempting to predict chemical longevity downhole. The effect that salinity has on the chemical kinetics is also significant, since to the authors' knowledge, there have been no previous reports on the effect of salinity on aldol condensations. (As it was a depressive effect and therefore industrially useless, this is not entirely surprising.) A model was built to predict the chemical degradation of GA within the temperature, pH, and salinity parameters tested and was shown to be consistent with two mixed-parameter scenarios, showing the kinetic influences do indeed have an additive combinatorial

effect. Whereas both pH (in the form of $[H^+]$) and temperature combine naturally in the Arrhenius equation, the effect of salt on the chemical kinetics is more nebulous. Should a secondary salt effect be at work and salinity is directly affecting the pH of the solution (as is most likely the case given the results of chapter four), it is perfectly sensible that this effect would be additive, inserting naturally into the Arrhenius equation, after which a true chemical kinetic model could be built therefore revealing the exact relationship between $[NaCl]$ and aldol condensation kinetics.

Future research on this topic is needed to elucidate the true effect that NaCl has on the chemical kinetics of aldol condensation. Because salt effect chemistry has been largely forgotten in recent chemical history and is extremely relevant to the downhole conditions of hydraulic fracturing, this is once again an important and interesting topic which warrants investigation. Furthermore, it has been shown that in S_N2 reactions of alkyl halides that at a concentration of .06 M, Cl^- begins to compete with water as the nucleophile in solution. This means, in theory, there may be some concentration at which Cl^- becomes more nucleophilic than the α -carbon nucleophile in the aldol condensation, leading to the formation of chlorinated byproducts instead of the relatively harmless GA polymer. Because $[Cl^-]$ levels can reach saturation, especially in saline shales such as the Marcellus, this is an extremely pertinent question which warrants further attention.

Chapter four expanded upon the experiments performed in chapter three to include the surfactant class NPEs. This research unequivocally shows that under the anaerobic, anoxic, hot, and mildly acidic environment created within hydraulic fracturing simulation vessels, NPEs break down via hydrolysis into the endocrine-disrupting compound NP (nonylphenol). This marks the first time that hydrolysis of NPE into NP has been shown to occur in a naturally-

relevant environment, mostly because the downhole environment of hydraulic fracturing is rather extreme and quite dissimilar from surface-level conditions. Conversely, this means the unique conditions provided by hydraulic fracturing are the *only* “natural” conditions capable of inducing hydrolysis of NPE into NP; it took days of “downhole conditions” (high heat, pressure, with mild acidity- pH 5- and low salinity- 0.5 M) before NP was detected via ToF-MS, an extremely unlikely scenario in the known environment in anything other than hydraulic fracturing (where this is actually commonplace and considered “mild” conditions). Just as was discovered in chapter three, salinity in levels that are commonplace in downhole environments had a drastic effect on chemical kinetics. This time, however, salinity accelerated the kinetics of the breakdown of the selected chemical- not surprising since hydrolysis in this instance was acid-catalyzed (whereas aldol condensation is the reverse, catalyzed by base). Because extremely saline environments often require the use of stronger surfactants such as NPE to solubilize other nonpolar constituents, as well as the increased need for corrosion inhibition which NPE also provides, the effect which salt has on the hydrolysis of NPE is especially worrisome. And though NP and NPE both exhibit strong sorptive effects, colloidal transport on small shale particles may enable NP to be mobilized with flowback water to escape downhole confines and become an environmental concern.

Future research on this subject is sorely needed because of the nature of NP’s toxicity; endocrine-disrupting compounds can have disastrous effects, especially on the unborn, even in small amounts. Furthermore, NP is highly bioaccumulative, increasing the potential hazard of even the slightest exposure. The colloidal transport of this chemical on shale particles should be heavily investigated because the solid waste from hydraulic fracturing (with which this shale sediment would end up) is not treated as toxic, and is generally disposed of in municipal

landfills. Also, as mentioned earlier, Cl^- in high concentrations can outcompete water as the “best” nucleophile in solution.²⁵⁸ Therefore this possibility should be investigated to determine the likelihood of chlorinated byproduct formation from the degradation of NPE.

In general, many other chemicals used in hydraulic fracturing can be tested in the same way that GA and NPE were with the framework laid out by the above research. Many biocides such as TPHS and DMO are known to degrade into formaldehyde, something that could be quickly proven to also occur downhole in the downhole simulation reactors. DBNPA and other biocides suspected to degrade into harmful nitrosamines can also be tested: because the downhole environment is so unlike surface conditions, many reactions which may have taken too long to be of any concern are drastically accelerated, suddenly making them relevant within the context of hydraulic fracturing. Truly, this shows the need for detail-oriented research prior to claims of safety when it comes to large-scale processes which occur in outdoor environments such as hydraulic fracturing. This research perhaps alludes to the fact that, once again, the long-term cost of the damage induced by industrial short-sightedness may far outweigh any short-term profit that was gained. Hopefully this body of research may help prevent the same mistake from being repeated many more times in the future.

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