

# **A REVIEW OF BIOFOULING CONTROLS FOR ENHANCED IN SITU BIOREMEDIATION OF GROUNDWATER**

---

**October 2005**

## TABLE OF CONTENTS

	<b>Page</b>
1.0 INTRODUCTION .....	1
2.0 BIOFOULING CAUSES AND MECHANISMS .....	3
2.1 CAUSES OF BIOFOULING.....	3
2.2 BIOFILM FORMATION .....	3
2.3 BIOFOULING OBSERVED IN EISB APPLICATIONS.....	4
3.0 WELL REHABILITATION VERSUS PREVENTATIVE CONTROL.....	11
3.1 WELL REHABILITATION.....	11
3.1.1 Physical Displacement Methods .....	11
3.1.2 Chemical Treatment .....	11
3.1.3 Other Rehabilitation Methods.....	12
3.1.4 Advantages and Disadvantages of Well Rehabilitation.....	12
3.2 PREVENTATIVE BIOFOULING CONTROLS .....	13
3.2.1 Biofouling Controls in Industrial and Water Treatment Applications .....	13
3.2.2 Nontoxic Coatings to Prevent Biofouling .....	14
3.2.3 Biofouling Controls in Groundwater Remediation Applications .....	15
3.2.3.1 Chlorine Dioxide.....	15
3.2.3.2 Tetrakis(hydroxymethyl) phosphonium sulfate (THPS)— Tolcide .....	15
3.2.3.3 Aqua Gard™ Process.....	17
3.2.3.4 Hydrogen Peroxide .....	17
3.2.3.4.1 Site 17, Robins AFB, Georgia .....	17
3.2.3.4.2 Site 19, Edwards AFB.....	18
3.2.3.5 Impressed Current Systems.....	18
3.2.3.6 Ultrasonic Tube Resonator.....	19
3.2.3.7 Summary.....	19
4.0 REVIEW AND INITIAL EVALUATION OF BIOFOULING CONTROLS .....	21
4.1 CHARACTERISTICS OF IDEAL BIOFOULING CONTROLS FOR EISB .....	21
4.2 IDENTIFICATION OF BIOFOULING CONTROLS .....	21
4.3 INITIAL EVALUATION AND SCORING OF BIOFOULING CONTROLS ...	30
5.0 DETAILED EVALUATION OF PROMISING BIOFOULING CONTROL OPTIONS 35	
5.1 CHLORINE DIOXIDE.....	35
5.1.1 Mode of Action.....	35
5.1.2 Generation of Chlorine Dioxide.....	36
5.1.3 Where Used.....	36
5.1.4 Impacts to Water Quality.....	38
5.1.5 Implementation Issues.....	38
5.2 SODIUM HYPOCHLORITE.....	38
5.2.1 Mode of Action.....	38
5.2.2 Where Used.....	39

## TABLE OF CONTENTS (continued)

	<b>Page</b>
5.2.3 Impacts to Water Quality .....	39
5.2.4 Implementation Issues .....	39
5.3 GLYCOLIC ACID.....	39
5.3.1 Mode of Action.....	39
5.3.2 Where Used.....	40
5.3.3 Impacts to Water Quality .....	40
5.3.4 Implementation Issues .....	40
5.4 AQUA GARD™ PROCESS.....	40
5.4.1 Mode of Action.....	40
5.4.2 Where Used.....	40
5.4.3 Impacts to Water Quality .....	40
5.4.4 Implementability Issues .....	40
5.5 THERMAL PASTEURIZATION .....	41
5.5.1 Mode of Action.....	41
5.5.2 Where Used.....	41
5.5.3 Impacts to Water Quality .....	42
5.5.4 Implementability Issues .....	42
6.0 CONCLUSIONS.....	43
7.0 REFERENCES .....	45

## LIST OF FIGURES

	<b>Page</b>
Figure 1	Schematic of Biofilm Formation. .... 5
Figure 2	Photographs of Fouling Related to Nutrient Delivery. .... 9
Figure 3	Performance of Chlorine Dioxide for Controlling Biofouling. .... 16
Figure 4	Schematic of Aqua Gard™ System. .... 41

## LIST OF TABLES

	<b>Page</b>
Table 1	Summary of Impacts of Biofouling on Selected Bioremediation Field Demonstrations ..... 6
Table 2	Typical Biofouling Controls Used in Industrial and Water Treatment Processes. 14
Table 3	Identification of Available and Emerging Biofouling Controls. .... 22
Table 4	Evaluation and Scoring of Biofouling Controls..... 31
Table 5	Cost Comparison of Biofouling Controls. .... 33
Table 6	Proposed Primary and Secondary Biofouling Controls for Field Demonstration/Validation. .... 36
Table 7	Commercial Chlorine Dioxide Generation Systems. .... 37

## ACRONYMS AND ABBREVIATIONS

---

AFB	Air Force Base
BCHT	blended chemical heat treatment
CDG	chlorine dioxide generator
CFB	Canadian Forces Base
ClO <sub>2</sub>	chlorine dioxide gas
DCE	dichlorethene
DoD	Department of Defense
EISB	enhanced in situ bioremediation
EPA	Environmental Protection Agency
EPS	exopolysaccharides
ESTCP	Environmental Security and Technology Certification Program
EtOH	ethanol
FIFRA	Federal Insecticidal, Fungicidal and Rodenticidal Act
gpd	gallons per day
gpm	gallons per minute
gpm/ft	gallons per minute per foot
H <sub>2</sub> O <sub>2</sub>	hydrogen peroxide
HOCl	hypochlorous acid
igpm	imperial gallons per minute
LBA	liquid biofouling agent
MeOH	methanol
mg/L	milligrams per liter
MMO	mixed metal oxide
NaClO <sub>2</sub>	sodium chlorite
NaOCl	sodium hypochlorite
NSF	(formerly) National Sanitation Foundation
O&M	operation and maintenance
ORP	oxidation-reduction potential
PCE	tetrachloroethene
PVC	polyvinyl chloride
RDX	hexahydro-1,3,5-trinitro-1,3,5-triazine

## ACRONYMS AND ABBREVIATIONS (continued)

---

SLN	special local needs
H <sub>2</sub> NO <sub>3</sub> S	Sulfamic acid
TCE	trichloroethene
TDS	total dissolved solids
TEX	toluene, ethylbenzene, and xylene
THPS	tetrakis(hydroxymethyl) phosphonium sulfate
TWA	time weighted average
UIC	user identification code
USACE	U.S. Army Corps of Engineers
UTR	ultrasonic tube resonator

*This page left blank intentionally.*

## 1.0 INTRODUCTION

Enhanced in situ bioremediation (EISB) is increasingly being used for the remediation of chlorinated solvents and recalcitrant chemicals (e.g., perchlorate, nitrate, hexahydro-1,3,5-trinitro-1,3,5-triazine [RDX]) in groundwater, both for plume containment and source area treatment. Given the dimensions (width, depth) of plumes and source areas at many Department of Defense (DoD) sites, EISB often requires extraction of impacted groundwater, amendment with soluble nutrients (electron donors or acceptors), and recharge of the nutrient-amended water to the aquifer to effectively mix and distribute the nutrients throughout the target treatment area. Unfortunately, the nutrient-rich conditions created within an injection well and the surrounding filter pack often favor rapid microbial growth and biofilm formation, which can result in a loss of well efficiency. When well efficiency declines below an acceptable level, physical well rehabilitation coupled to aggressive chemical shock treatment is typically required to restore the use of the well for continued nutrient delivery. Physical well rehabilitation processes are labor-intensive and costly, particularly when frequent well rehabilitation is required. In fact, well rehabilitation can be the most significant operating cost at EISB sites (McCarty et al, 1998) and can reduce the cost-effectiveness of an EISB approach to the point that more conventional remedial approaches (e.g., pump-and-treat) have lower operation and maintenance (O&M) and overall life-cycle costs. Accordingly, lower-cost, effective biofouling controls that prevent the occurrence of this problem are required.

The objective of this report is to review well rehabilitation and biofouling controls that are potentially relevant to EISB applications and to identify promising biofouling controls for comparative field evaluation and validation under Environmental Security and Technology Certification Program (ESTCP) Project ER-0429. The field demonstration will evaluate and compare the performance of multiple preventative biofouling controls for nutrient delivery wells used in EISB applications and will generate cost and performance data for biofouling controls that can be readily implemented at DoD facilities with varying geochemistry and infrastructure. The information generated from the project will also be used to develop technical guidance to assist DoD remediation project managers and practitioners with selection and deployment of successful preventative biofouling controls for EISB projects.

The remainder of this review presents a summary of biofouling causes and mechanisms (Section 2); a discussion of the differences between well rehabilitation and preventative biofouling control, including a review of case studies where biofouling controls have been used in groundwater remediation applications (Section 3); identification, evaluation and scoring of promising biofouling control options for further field evaluation/validation (Sections 4 and 5); and conclusions for preventive biofouling controls (Section 6).



*This page left blank intentionally.*

## **2.0 BIOFOULING CAUSES AND MECHANISMS**

To evaluate biofouling controls, it is important to understand the causes and mechanisms of biofouling in EISB applications. In the following sections, the causes of biofouling, biofilm formation, and field system observations of biofouling are reviewed.

### **2.1 CAUSES OF BIOFOULING**

Biofouling occurs when bacteria attach, grow and block the well screen, filter pack, or formation surrounding a nutrient delivery well, thereby limiting or preventing the proper function of the well. The bacteria may originate in the aquifer itself or may be introduced during well installation, amendment addition, or groundwater recirculation (Cullimore, 1999). Most groundwater environments contain an active and diverse microbial population, but growth is limited in the absence of high concentrations of nutrients (Fry et al, 1997).

Bacterial growth within delivery wells is generally accelerated by adding the provided nutrients to promote the desired EISB reactions. For EISB applications at sites impacted by chlorinated solvents, nitrate, perchlorate, and/or RDX, these nutrients typically consist of carbon-based electron donors (e.g., sugars, alcohols, organic acids, edible oils) or hydrogen. In these cases, the contaminants serve as electron acceptors, and their reduction (respiration) is linked to metabolism of the electron donors. While biofouling of the wells may occur by bacteria using the electron donors to degrade the target contaminants, it may also result from bacteria that are using oxygen, nitrate, iron, manganese, or sulfate as electron acceptors coincidentally with degradation of the target contaminants. Essentially, the electron donors being added can promote a wide range of microbial metabolic activity that can cause biofilm formation and well biofouling. For EISB applications at sites impacted by petroleum hydrocarbons, fuel oxygenates, and/or ketones, added nutrients typically consist of electron acceptors such as oxygen, nitrate, or sulfate. Microbial growth and biofouling are generally more rapid under aerobic conditions due to the higher growth yields of microorganisms that respire oxygen. Nitrate- and perchlorate-reducing bacteria also have high growth rates and have been found to cause significant fouling of nutrient delivery wells.

Factors affecting the growth of microorganisms in nutrient delivery wells include the carbon:nitrogen:phosphorus ratio, temperature, redox potential, and pH of the aquifer (Cullimore, 1999). In EISB applications, groundwater temperatures are moderate (generally between 12 and 25°C, depending on location and season), which tends to accelerate microbial growth. Because most bacteria grow within a pH range of 5 to 9, it stands to reason that biofouling will be most problematic within this pH range. The presence of significant fines and elevated concentrations of divalent cations (e.g., calcium and magnesium) in groundwater/fluids being injected via delivery wells can also exacerbate biofilm formation and accelerate well fouling, as discussed further in Section 2.2.

### **2.2 BIOFILM FORMATION**

The process of biofilm formation and biofouling is a multistep process. First, most bacteria prefer to be attached to a surface, and the aquifer provides very high surface area and a positively charged surface for bacterial attachment. Bacteria adhere to the porous media through

electrostatic interactions, hydration forces, and hydrophobic and steric interactions (Elimelech et al., 1995). Bacterial adhesion may be influenced by factors such as mineral and bacteria surface charges, mineral surface roughness, exopolymer formation, predation, and competition. Bacteria also adhere to well screen materials and the filter-pack, which have lower surface-to-volume ratios than the surrounding aquifer materials, and so tend to foul quickly.

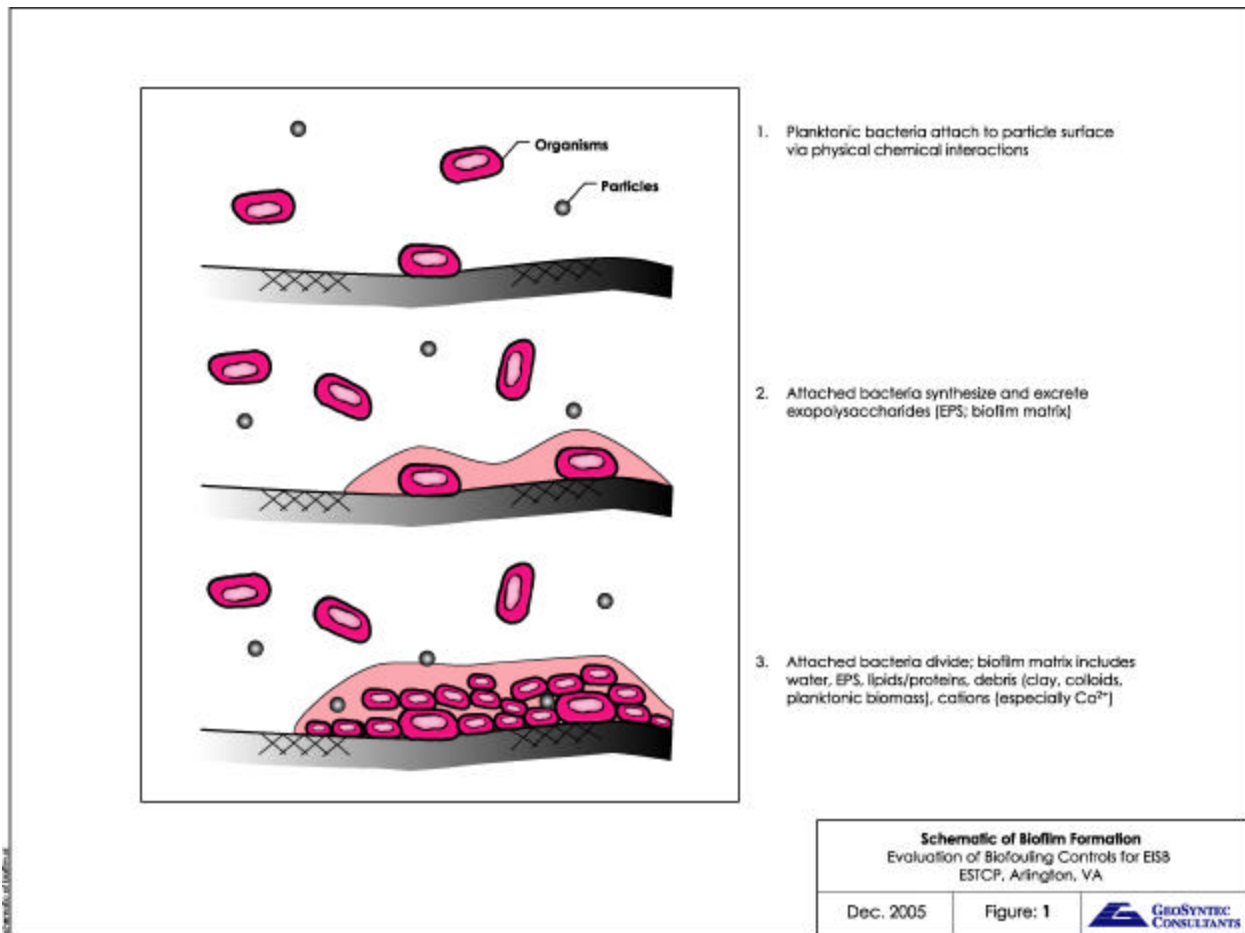
Once attached, the bacteria reproduce and grow to form colonies. The colonies grow outward to form a biofilm, by secreting exopolysaccharides (EPS), a water retaining matrix. The composition and structure of the EPS, as well as their physical and chemical properties, can vary widely and depend on the microbial community composition and the presence of ions in solution. Different microbial strains grow to form a community or consortial biofilm (Alford and Cullimore, 1999). A biofilm can be viewed as an organic polymer gel with living microorganisms trapped inside, as depicted in Figure 1. The nature of the gel can influence heat and mass transport properties, thus influencing the effectiveness of biocides (Characklis and Marshall, 1990).

The presence of multivalent cations (e.g.,  $Mg^{2+}$ ,  $Ca^{+2}$ ) enhances the mechanical strength of biofilms, enabling them to withstand significant fluid shear forces (Mayer et al, 1999). Biofilms can harden through the bioaccumulation of iron and other metallic cations such as Fe, Mn, Al, Cu, and Zn (Cullimore, 1999; Smith, 1995) and through the synthesis of crystalline structures usually based on carbonates. During this process, there is also the entrapment of clays, silts, and sands (USACE, 2000). In anaerobic biofilm systems, sulfate-reducing bacteria can generate hydrogen sulfide, which can react with iron and manganese to form metallic sulfides that can become trapped in the biofilm matrix. Methanogens can produce methane and carbon dioxide, which can also become trapped in the biofilm, or the gas alone can act to occlude pores in the aquifer matrix (Cullimore, 1999).

Due to fluid shear in the aquifer, attached biomass can be dislodged from the biofilm and transported by the water phase to another location, where it can re-attach. This sloughing/re-attachment can substantially redistribute biomass in porous media (Taylor and Jaffe, 1991). Eventually, the biofilm becomes sufficiently enlarged that it occupies a significant fraction of the void volume of the well screen, filter pack, and/or aquifer, and occlusion or fouling occurs (Cullimore, 1999), resulting in decreased well performance and reduced EISB effectiveness.

### **2.3 BIOFOULING OBSERVED IN EISB APPLICATIONS**

As a result of the relatively recent interest in enhanced bioremediation, reliable data on the frequency of biofouling problems is primarily limited to anecdotal evidence from field-scale bioremediation studies that have been completed to date. As previously indicated, EISB strategies typically involve either the addition of an organic substrate or hydrogen as an electron donor (e.g., to promote contaminant reduction) or an inorganic electron acceptor (e.g., oxygen for aerobic oxidation of petroleum hydrocarbons). Nutrient addition creates environmental conditions favoring rapid microbial colonization and growth, especially in the immediate vicinity of the well screen and filter pack. Accordingly, nutrient amendment schemes that result in nearly continuous nutrient availability within the filter pack are more likely to experience biofouling problems. As shown in Table 1, 17 of 20 bioremediation studies surveyed reported some degree of biofouling. Interestingly, several of the studies where biofouling was



**Figure 1. Schematic of Biofilm Formation.**

not experienced, added nutrients infrequently (i.e., less frequently than weekly). In the absence of proven biofouling controls applicable to in situ remediation, rehabilitation technologies, rather than preventative measures, have been typically utilized in response to significant biofouling (i.e., as measured by rapid loss of well performance). Consequently, the biofouling controls most commonly employed in these studies include conventional swabbing/surging and/or shock chlorination.

Photographs of biofouling related to several EISB applications are provided in Figures 2a and 2b. Figure 2a provides comparative photographs (from down-hole video) of unfouled and fouled sections of the well screen of an electron donor delivery well used for perchlorate remediation. Figure 2b shows biofouling of an extraction well pump from a recirculation-based EISB application where electron donor was not completely metabolized before reaching the extraction well. In both cases, rehabilitation of the well/equipment was required to maintain EISB operations.

**Table 1. Summary of Impacts of Biofouling on Selected Bioremediation Field Demonstrations**

<b>Contaminant</b>	<b>Location</b>	<b>Reference/Source</b>	<b>Nutrient Amendment Approach</b>	<b>Impact on Well Performance</b>	<b>Fouling Control/Response</b>
Trichloroethene (TCE)	Dover Air Force Base (AFB), DE	Ellis et al (2000)	Continuous recirculation with lactate (100 milligrams per liter [mg/L]), ammonia, and phosphate	Severe loss of injection well efficiency	Physical rehabilitation (brushing and overpumping) combined with hypochlorite addition
TCE	Cape Canaveral AS, Florida	GeoSyntec	Pulsed injection (1 hr per day) of 520 mg/L ethanol (EtOH) (time weighted average [TWA] concentration)	Severe loss of injection well efficiency causing system shutdowns	Physical rehabilitation (surging and overpumping)
TCE	Edwards AFB, California	McCarty et al (1998)	Injection of oxygen (up to 44 mg/L) and toluene (up to 13.4 mg/L)	Loss of injection well efficiency	Physical rehabilitation and pulsed addition of peroxide at concentrations up to 117 mg/L
TCE	Industrial facility, ON, Canada	GeoSyntec	Pulsed injection (1 hr per day) of 500 mg/L TWA of EtOH or methanol (MeOH)	Loss of injection well efficiency	Physical rehabilitation (brushing and overpumping)
TCE	Industrial facility, California	Beak Consultants	Pulsed injection (1 hr per day) of 1,000 (TWA) mg/L of MeOH	Severe loss of injection well efficiency causing system shutdowns	Periodic liquid biofouling agent (LBA) cleaning events, surging and overpumping, and initiation of daily pulses of hydrochloric acid following donor injection (marginal improvement)
TCE	Industrial facility, Pennsylvania	O'Brien & Gere/GeoSyntec	Daily pulse injection of EtOH (50 mg/L TWA)	Impacts to delivery and extraction well efficiency	Physical rehabilitation (brushing and overpumping)
TCE	Industrial facility, Massachusetts	GeoSyntec	Weekly addition of acetate and MeOH at 100 mg/L and 500 mg/L TWA, respectively	Significant loss of well efficiency due to biological clogging of the well screen, causing automatic shut-down of the system	Injection well cleaned every 3 months using glycolic acid (i.e., LBA), surging and overpumping
Tetrachloroethene (PCE)	Kelly AFB, Texas	Major et al(2002)	Daily pulsed injection of MeOH and acetate (3.6 mM each)	Some loss of injection well efficiency and biofilm in aboveground process piping	Rehabilitation with hypochlorite and overpumping

**Table 1. Summary of Impacts of Biofouling on Selected Bioremediation Field Demonstrations (continued).**

Contaminant	Location	Reference/Source	Nutrient Amendment Approach	Impact on Well Performance	Fouling Control/Response
PCE	Dover AFB, Delaware	GeoSyntec	Daily injection of EtOH (63 mg/L TWA) and lactate (30 mg/L TWA)	Biofilm observed on aboveground process piping and higher water levels in injection well	Some loss of well capacity; implemented physical rehabilitation (brushing and overpumping) and hypochlorite for above-ground piping
PCE	Industrial Facility, South Carolina	GeoSyntec	Daily pulse injection of MeOH (100 mg/L TWA) and lactate (25 mg/L TWA)	Biofilm observed on aboveground process piping and impacts to delivery and extraction well efficiency	Physical rehabilitation (brushing and overpumping) and hypochlorite for above-ground piping
PCE	Bachman Road Residential Wells Site, Michigan	Lendvay et al (2003)	Continuous recirculation with lactate (0.1 to 1.0 mM) with phosphate and nitrate	Loss of well efficiency	Control by minimizing donor dosing and frequent jetting of the injection wells
Carbon Tetrachloride	Schoolcraft, Michigan	Dybas et al (1998)	Natural gradient flow with periodic acetate amendment (100 mg/L) and pH adjustment to 8.3	None documented	Controlled using pulsed donor delivery (6 hr per wk), alternate use of wells as either injection or extraction, and reversal of groundwater flow for one hr following donor amendment
Carbon Tetrachloride	Canadian Forces Base (CFB) Borden, ON, Canada	Devlin and Muller (1999)	Semipassive pulsed acetate amendment every 5 weeks	None observed after 1.3 yr of operation; biofilm observed on the interior surface of injection well casings	Intermittent donor additions
Perchlorate	Industrial facility, Nevada	GeoSyntec	Daily injection of EtOH or citric acid in high total dissolved solids (TDS) groundwater	Severe loss of well efficiency due to biofouling and mineral precipitation with EtOH; only biofouling with citric acid	pH shift with citric acid prevented mineral precipitation; chlorine dioxide prevented biofouling
Perchlorate	Rocket facility, Nevada	GeoSyntec	Daily injection of citric acid	Loss of well efficiency due to biofouling	Reduce injection flow rate
Perchlorate	Aerojet Superfund site, California	Cox et al (2002a)	Recirculation with pulsed acetate or lactate addition (TWA 50 mg/L)	Loss of well efficiency due to biofouling	Reduce injection flow rate

**Table 1. Summary of Impacts of Biofouling on Selected Bioremediation Field Demonstrations (continued).**

Contaminant	Location	Reference/Source	Nutrient Amendment Approach	Impact on Well Performance	Fouling Control/Response
Perchlorate	Aerojet Superfund site, California	Cox et al (2002a)	Single-pass active biobarrier with pulsed EtOH addition (TWA 50 mg/L)	Loss of well efficiency due to biofouling prior to use of chlorine dioxide for biofouling control	Reduced injection rate, rate sustained following implementation of daily chlorine dioxide pulses
Perchlorate	Aerojet Superfund site, California	Cox et al (2002a)	High volume recharge (100 to 150 gallons per minute [gpm]) with pulsed EtOH, acetate or citric acid addition (30 mg/L TWA)	Loss of well efficiency and system shutoff when chlorine dioxide not employed	Chlorine dioxide effectively maintained well efficiency for 3-mo operation period
Nitrate	Research site, New Mexico	Eric Nuttall, University of New Mexico	Electron donor addition via five-spot well configuration	Loss of well efficiency due to biofouling	Periodic peroxide doses (limited performance improvement)
Cl-ethenes, Cl-ethanes, ketones, TEX	Farmington, New Hampshire	Cox et al (2002b)	Continuous addition of 400 mg/L sulfate via three wells in gravel delivery trench	Seasonal biological/mineral fouling decreased efficiency of injection wells causing excessive mounding in trench	Injection wells were cleaned once with LBA surging and overpumping. The wells returned to near their original yields following treatment.

*Notes:*

TCE - trichloroethene

EtOH - ethanol

PCE - tetrachloroethene

MeOH - methanol

TEX - toluene, ethylbenzene, and xylene

TWA - time-weighted average

2a) Comparative photos (from down-hole video) of unfouled and fouled sections of the well screen



Unfouled Well Screen



Fouled Well Screen

2b) Biofouling of an extraction well pump from a recirculation-based EISB application



Photographs of Fouling Related to Nutrient Delivery  
Evaluation of Biofouling Controls for EISB  
ESTCP, Arlington, VA

Dec. 2005

Figure: 2



Figure 2. Photographs of Fouling Related to Nutrient Delivery.



*This page left blank intentionally.*

## **3.0 WELL REHABILITATION VERSUS PREVENTATIVE CONTROL**

Groundwater remediation practitioners typically rehabilitate wells in response to the occurrence of significant biofouling rather than using biofouling controls in a preventative manner. In the following sections, conventional well rehabilitation is discussed and contrasted to preventative biofouling controls used in industrial and groundwater applications.

### **3.1 WELL REHABILITATION**

Well rehabilitation is employed in response to significant biofouling and most commonly involves physical swabbing and pumping procedures, coupled with the use of aggressive and potentially hazardous chemicals. Heat may also be used to augment the performance of biocidal chemicals. Rehabilitation is recommended if the well yield, efficiency, or specific capacity declines by more than 25% (ADITC, 2002), but due to the cost of these procedures, well rehabilitation is often initiated only when well yields decline by 50 to 75% (GeoSyntec experience). In this section, physical displacement methods, chemical treatments, and other rehabilitation methods are discussed.

#### **3.1.1 Physical Displacement Methods**

Surging with overpumping is a common well rehabilitation procedure. Surging can be performed by using surge blocks or by injecting air in the casing above the well screen. It is labor-intensive and often requires specialized equipment (e.g., service rigs). Manual brushing is also effective in dislodging material from the well screen and casing (Smith, 1995). Over-pumping involves removing water from the well, either by bailing or pumping, and allows water from the aquifer to flow into the well, removing any fines or biofilm fragments that were dislodged through surging or brushing.

Jetting approaches may also be used to dislodge fines and biofilms from well screens. Jetting is carried out using a perforated jetting tool and a high-pressure water source. Because jetting has the potential to pack debris against the borehole wall, it is coupled with an airlift pump to promptly remove the debris (Smith, 1995).

#### **3.1.2 Chemical Treatment**

Historically, shock chlorination has been used to prevent biofouling. Chlorine (e.g., bleach) is added at concentrations in the 500 to 2,000 mg/L range and generally precedes acid treatment. After treating for 24 hr, the chlorinated water is surged within the well and pumped out. Purge water with any chlorine residue is pumped to open retention ponds or tanks to allow the chlorine to dissipate prior to discharge to a wastewater treatment facility. Use of chlorine can result in the formation of disinfection byproducts (e.g., trihalomethanes) through reaction with natural organic carbon or other organic compounds present in the groundwater.

Muriatic acid (industrial grade hydrochloric acid), sulfamic acid, and glycolic acid are also commonly used for well rehabilitation (ADITC, 2002). Acids are used to dissolve iron and manganese oxides and carbonate encrustation and exert an antibacterial effect by providing a pH shock to bacteria typically adapted to neutral pH (Smith, 1995).

Muriatic acid is a powerful acid and is most effective for the removal of mineral scale. It is hazardous to handle, requiring field personnel to wear full-body splash protection and respirators as it can generate toxic fumes. Muriatic acid can also be contaminated with trace levels of arsenic and other metals (undesirable for introduction to groundwater environments) and poses purge water handling problems due to its low pH (Smith, 1995). In contrast to muriatic acid, sulfamic acid comes as a solid, which is stable and relatively safe to handle and mix; however, it can form ammonia upon dissolution (Smith, 1995). Glycolic acid, also known as hydroxyacetic acid, is a liquid organic acid, commercially available in 70% concentrations as LBA (CETCO, Arlington Heights, Illinois). It is safer to use than sulfamic and muriatic acids, being noncorrosive and producing little or no toxic fumes (ADITC, 2002). Glycolic acid has antibacterial and metal chelating properties and is particularly suited to attacking iron bacteria biofilms. Being weaker than sulfamic acid, longer contact times are required (ADITC, 2002), which can translate into longer EISB system shutdowns and higher O&M costs.

After an acid is added to a well, water is added to the well to push the acid solution through the screen and into the filter pack and formation immediately surrounding the well. The acid solution is mechanically agitated, left in the well to react with encrustations and biofilms, agitated again, and then pumped to waste. The treatment time varies from a few hours to more than 15 hr, depending of the severity of the fouling and the type of acid used. Acidic purge water requires neutralization prior to being pumped to a wastewater treatment facility or containerized and disposed of in an environmentally safe manner (ADITC, 2002).

### **3.1.3 Other Rehabilitation Methods**

Hot water (54°C or 130°F) has been used to augment or replace chemical treatment to kill and disperse iron bacteria in wells. However, heat may enhance bacterial growth away from the thermal shock area, resulting in fouling within the aquifer itself. Heat can also cause shrinking of bentonite grout, adversely affecting well integrity (ADITC 2002; Alford and Cullimore, 1999). Other well rehabilitation technologies include Aqua Freed<sup>TM</sup>, which involves the injection of cryogenic carbon dioxide (Mansuy, 1999), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) (Smith, 1995), and blended method treatments, such as the blended chemical heat treatment (BCHT) process (developed by ARCC, Inc., Daytona Beach, Florida, U.S. Patent # 4,765,410) that incorporates physical, chemical, and heat treatments.

### **3.1.4 Advantages and Disadvantages of Well Rehabilitation**

The advantages of conventional well rehabilitation procedures include restoration of injection well performance (although this is usually temporary) using methods that are relatively straightforward and widely available. For shallow sites (e.g., less than 20 ft deep), it may be possible to maintain delivery well capacity through simple and cost-effective brushing, surging, jetting and/or overpumping techniques (with or without added chemicals) as part of a prescribed O&M program.

The disadvantages of well rehabilitation include the cost, requirement for process shutdown, and, in some cases, transient improvement in well performance. For example, in a U.S. Army Corps of Engineers (USACE) Repair, Evaluation, Maintenance, and Rehabilitation study evaluating potentially cost-effective rehabilitation techniques for relief wells at the Leesville Dam in

Leesville, Ohio, the rehabilitation procedure employed polyphosphate addition, surging (2 to 4 hr), and shock-chlorination (1,000 mg/L for 12 hr), followed by well redevelopment using surging and over-pumping (Alford and Cullimore, 1999). While this was highly effective in restoring well performance, the application of such intensive rehabilitation measures on a frequent basis (e.g., monthly or quarterly) would increase the operating cost of an EISB treatment system to the point that the technology might not be cost-effective relative to conventional remediation technologies (e.g., pump-and-treat).

At many EISB sites, well rehabilitation can be the most significant operating cost (McCarty et al, 1998). Well rehabilitation has been estimated to cost in the vicinity of \$3,000 to \$12,000 per well (depending on well diameter, depth, and degree of fouling), when subcontractor, contractor oversight, decontamination and purge water treatment costs are included (Smith, 1995). One of the greatest indirect costs of well fouling is the loss of well and process efficiency (Smith, 1995). For example, plugging increases the energy burden of the pump to move the same volume of water (Helweg et al, 1983), and biofilms immediately surrounding or fouling injection wells can increase nutrient consumption.

Rehabilitation is often only partially successful. In ideal cases, the well may remain unclogged for years. However, it is much more common that performance is maintained only for weeks to months (Smith, 1995).

## **3.2 PREVENTATIVE BIOFOULING CONTROLS**

Preventative biofouling controls used in industrial or groundwater applications typically attempt to inhibit or inactivate the bacterial populations forming biofilms through the use of oxidizing biocides or concentrated acids; displace the biomass by physical means; and/or destabilize the biofilm matrix using surfactants, dispersing agents, or chelating agents. Ideally, biofouling controls would prevent the formation of biofilms in both the well screen and filter pack, eliminating or minimizing the need for well rehabilitation. Biofouling controls that can be automated to eliminate labor and downtime associated with well rehabilitation will reduce the cost associated with EISB applications.

### **3.2.1 Biofouling Controls in Industrial and Water Treatment Applications**

Biofouling controls are used in a wide array of industrial and water treatment applications, as detailed in Table 2. Typical applications include heat exchange and cooling systems, food processing operations, medical and dental equipment cleaning systems, and water treatment distribution systems. A variety of biofouling controls, such as nonoxidizing biocides, physical methods, surfactants, enzymes, and acids are employed.

Industrial systems differ significantly from typical environmental applications, which limit the applicability of many industrial controls for use as EISB biofouling controls. Often, industrial equipment is readily accessible to physical cleaning, whereas the filter pack and/or the surrounding formation in EISB systems are not readily accessible and have the potential to biofoul significantly. Industrial systems are typically closed systems, and as a result, there is minimal risk of adverse environmental impacts resulting from exposure to process water containing a toxic anti-fouling agent which may not be desirable for release into groundwater or

**Table 2. Typical Biofouling Controls Used in Industrial and Water Treatment Processes.**

<b>Process</b>	<b>Biofouling Control Process</b>	<b>Reference</b>
Heat exchange systems	Nonoxidizing biocides, physical (e.g., brushing, pigging), surfactants, and biocidal coatings	Cloete et al (1992)
Food processing equipment	Enzymes, acids and bases, surfactants, and oxidizing biocides	Parkar et al (2004)
Medical/Dental Implants and Infections	Antibiotics, heat sterilization, and oxidizing biocides	Neu et al (1992)
Marine coatings (hulls)	Inhibitory coatings	Zinn et al (2000)
Water Treatment <i>Filtration</i>	Physical (e.g., backwashing)	Montgomery (1985)
<i>Distribution</i>	Oxidizing biocides	Camper et al (1999)
<i>Nanofiltration</i>	Substrate (NOM) removal	Urfer et al (1997)
	Surfactants, acids and bases, and chelants	Liikanen et al (2002)
Wastewater infiltration	Oxidizing and nonoxidizing biocides	Baveye et al (1998)

Notes:

NOM – natural organic matter

drinking water supply aquifers. Finally, most industrial processes do not involve the beneficial use of bacteria as part of the process; all microbial activity is considered deleterious. Therefore, one can apply a biocide liberally without concerns about killing beneficial bacteria. In the case of an EISB system, one must balance control of bacterial activity in the delivery well, filter pack and immediate surroundings, while promoting a desired microbial activity (contaminant degradation) within a target treatment area.

### **3.2.2 Nontoxic Coatings to Prevent Biofouling**

Nontoxic coatings have been used by the Navy as a means of preventing biofouling of ships. For example, a nontoxic silicon fouling-release agent was developed by James R. Griffith for the U.S. Navy (patented 14 January 2004; Griffiths, 1995; 1997) and funded by ESTCP. In general, the fouling release coating is composed of a bonding layer and a release layer. The bonding layer is a one-component, nontoxic silicone rubber composed of organopolysiloxane and a polymeric toughening agent, and the release layer is nontoxic, liquid silicone rubber composed of organopolysiloxane containing terminal silicon-bonded hydroxyl groups, an alkyl silicate, and a curing agent. The bonding layer is applied to the unprotected material followed by the release layer. Both the bonding and release layers are liquids that cure at room temperature and can be applied to surfaces in the same fashion as common paints (brushing, spraying, dipping). The nonstick coating is meant to prevent adhesion of or fouling by organisms such as barnacles, tube worms, and algae.

Nonfouling coatings were evaluated for their likely effectiveness for biofouling control in bioremediation applications. Although the nonfouling coating may prevent or decrease the growth of a biofilm within the well screen itself, bacterial growth within the filter pack and surrounding aquifer materials are not likely to be affected. Furthermore, these coatings have largely been tested on barnacles and algae, with no data showing their effectiveness for microbial

fouling. The potential advantage of this approach is that, once cured, the coating is not anticipated to alter geochemistry or biodegradation mechanisms. Besides initial application of the coating by a licensed contractor, no further instrumentation or infrastructure is required. No handling issues have been identified for field practitioners, no regulatory issues anticipated, and no adverse water quality effects expected. Initial labor costs to apply the coating and any additional capital costs are anticipated to be low. However, operations and maintenance costs are expected to be high due to additional application of other biofouling controls to remove or reduce biofilm growth within the filter pack and surrounding aquifer matrix.

### **3.2.3 Biofouling Controls in Groundwater Remediation Applications**

Very few field studies exist that have assessed biofouling controls in groundwater remediation applications. The following sections summarize previous studies evaluating various types of biofouling controls.

#### **3.2.3.1 Chlorine Dioxide**

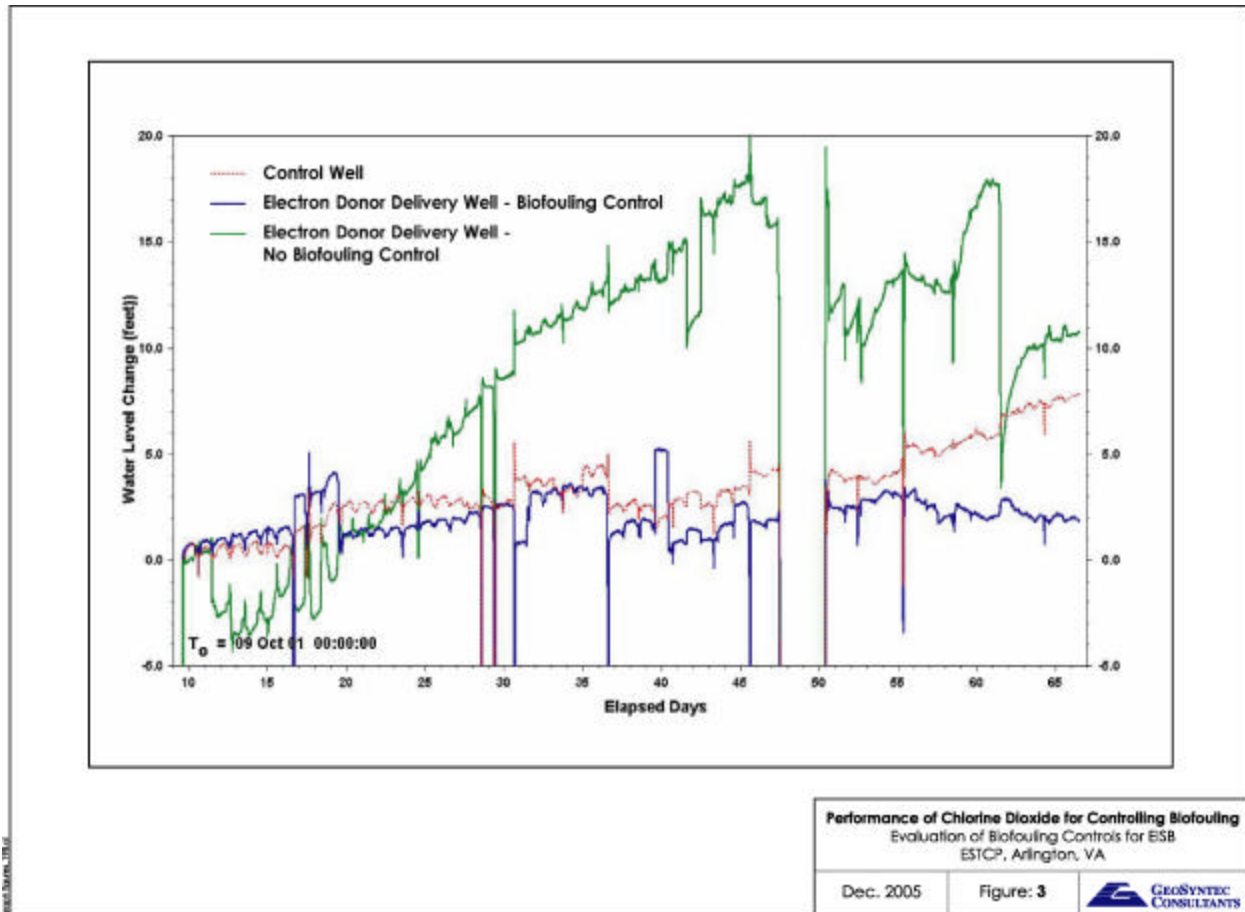
GeoSyntec has used chlorine dioxide gas ( $\text{ClO}_2$ ) to control biofouling in electron donor delivery wells at a number of sites employing EISB (GeoSyntec, 2003; 2004; Cox et al, 2003). Chlorine dioxide was generated using the chlorine dioxide generator (CDG) process, whereby a pre-blended pressurized mixture of nitrogen and chlorine gas (96% nitrogen, 4% chlorine) is passed through a cylinder of sodium chlorite ( $\text{NaClO}_2$ ) to generate 8%  $\text{ClO}_2$  in nitrogen. The  $\text{ClO}_2$  was piped directly into the recharge water in the injection well daily for 1 hr at a dose of 1 mg/L. Chlorine dioxide was effective in controlling biofouling in the electron donor delivery well over a sustained period of more than 6 mo. Figure 3 shows the difference in water levels for electron donor delivery wells, with and without chlorine dioxide for a period of 65 days. The water level in the electron donor delivery well that did not receive the chlorine dioxide rose almost 20 ft. The well that received chlorine dioxide exhibited water level changes of only a few ft, similar to wells that were not receiving electron donor (i.e., water level increases were due to local/regional groundwater elevation changes).

Another chlorine dioxide system was used to control biofouling in a perchlorate bioremediation system (ESTCP Project ER-0224). It was effective over a period of 6 mo until a valve malfunctioned, causing the water level to increase significantly and necessitating rehabilitation of the well.

#### **3.2.3.2 Tetrakis(hydroxymethyl) phosphonium sulfate (THPS)—Tolcide**

As part of a study evaluating the performance of vitamin B12-catalyzed reductive dechlorination at Aberdeen Proving Ground, Maryland, Millar et al, 2001, reported that a daily, 4-hr dose of 150 mg/L of THPS (a nonoxidizing biocide) inhibited biofouling around an injection well. Tolcide PS200 (20% THPS) was the commercial product employed.

The recirculation well was 10 in in diameter and consisted of two screened intervals separated by an inflatable packer. During Phase 1, groundwater was recirculated in the upper portion of the well and water levels increased 8.5 ft after 2 wk of injection of a vitamin B12, Ti(III)-citrate, and



**Figure 3. Performance of Chlorine Dioxide for Controlling Biofouling.**

glucose solution into the recirculation well, causing eventual shut-down of the system after 14 wk (Millar et al., 2001).

During Phase 2, ground water was recirculated through the bottom portion of the well, and Tolcide was added for 4 hr to achieve a concentration of 150 mg/L THPS. This study revealed that daily applications of Tolcide were successful in maintaining water levels relatively stable, with a rise of only 1 ft over a 12-wk period. When daily applications ceased, biological growth was immediate. Multiple weekly Tolcide soakings were used to restore well conditions after biofouling had occurred.

The addition of Tolcide during injection had a limited impact on the aquifer, and concentrations of Tolcide were low enough to not affect the continued biological degradation of cis- and trans-dichloroethene (DCE). Bacteriostatic activity was limited to the anaerobic treatment zone within 10-15 ft of the well and did not interfere with ongoing biological degradation outside this region.

Tolcide is a registered pesticide, which is not registered for use in groundwater. The use of Tolcide as a biofouling agent in groundwater requires the submittal of a special local needs (SLN) exemption permit under Section 24 (c) of the Federal Insecticidal, Fungicidal and Rodenticidal Act (FIFRA). The SLN must show that the use of Tolcide is required and an appropriate federally registered product is not available to perform the required task. The

environmental fate of THPS, the key ingredient in Tolcide, must be shown to not have adverse effect on humans or the environment at the planned application rate and concentration. The following specific details must be explained in the permit application:

- Site background
- Treatment system description
- Site specific information on groundwater characteristics
- An evaluation of the environmental fate of THPS in the groundwater at the site based on the results from modeling the fate in groundwater at the anticipated application rate and concentration, including predicted half-life of THPS, octanol-water partition coefficient, and anticipated concentrations of THPS in the groundwater.

More recently, Tolcide has been used for biofouling control at a full-scale EISB application at Dover AFB, Delaware. An SLN exemption, which took several years to procure, was obtained. Tolcide performance data from this application has yet to be reported in the scientific literature.

### **3.2.3.3 Aqua Gard™ Process**

Aqua Gard™ is a biofouling control process, which uses liquid and gaseous carbon dioxide to remove biofouling from water and groundwater wells. The injection of the carbon dioxide facilitates detachment of the biofilm in the well screen, and the surrounding filter pack and debris is removed using a dedicated pump or airlift. The use of the Aqua Gard™ process in a water supply well resulted in significant improvements in both well yield and water quality (Mansuy, 2003). The 6-inch diameter, 261-ft deep water supply well had experienced poor water quality and plugged within a month of operation due to the high organic content of the water. The Aqua Freed™ system was initially used to increase the capacity of the well. Following this treatment, the Aqua Gard™ system was installed to provide continuing biofouling control for the test well. The Aqua Freed™ initially increased the specific capacity of the well from 0.12 to 0.58 gpm/ft, while the continued monthly use of Aqua Gard™ increased the specific capacity to 0.61 to 0.86 gpm/ft. After 3 mo, the frequency of Aqua Gard™ application was decreased to every 2 mo. In summary, the Aqua Gard™ process appears to have promise for biofouling control; however no third party evaluations are currently available in the literature.

### **3.2.3.4 Hydrogen Peroxide**

#### *3.2.3.4.1 Site 17, Robins AFB, Georgia*

H<sub>2</sub>O<sub>2</sub> was used as a biofouling control for extraction wells at Site 17, Robins AFB. As a preventative measure, the recovery wells OT17EW1, OT17EW4 and OT17EW5 were each shocked with 5-, 10-, and 15-gal maintenance doses, respectively, of 50% H<sub>2</sub>O<sub>2</sub>, on December 11, 2003. Each well was 6 in in diameter. OT17EW1 and OT17EW4 each had 15 ft of screen and were 46 ft and 40 ft deep, respectively, while OT17EW5 had 50 ft of screen and was 110 ft deep. This treatment resulted in an initial improvement in well yield in wells OT17EW4 and OT17EW5. The daily flow total in well OT17EW4 improved from an average of 10,103 gallons



per day (gpd) in the 5 days prior to treatment, to an average of 11,013 gpd in the 5 days following treatment. OT17EW5 showed a similar but more dramatic increase from 49,500 gpd to 57,595 gpd. However, the flow rates in both wells slowly decreased with time, equilibrating at values similar to the initial flow rates in about a month. Interestingly, well OT17EW1 actually exhibited a decrease in daily flow from 8,422 gpd to 6,823 gpd after treatment.

Backflushing with 300 gal of water was performed as a maintenance treatment for extraction well OT17EW3 on December 11, 2003, but also resulted in a slight decrease in overall flow from 31,752 gpd to 30,077 gpd after treatment. This well was further treated with a rehabilitation fluid of 67% calcium hypochlorite (50 lb) mixed with 500 gal of water. The result was an increase in flow rate for the first 4 days after the start of pumping, but the flow rates trended lower in the months following rehabilitation.

In summary, H<sub>2</sub>O<sub>2</sub> and calcium hypochlorite treatment were effective but short-lived in their effectiveness for controlling biofouling.

#### *3.2.3.4.2 Site 19, Edwards AFB*

At Site 19, Edwards AFB, TCE-contaminated water was treated using an in situ aerobic cometabolic biodegradation system (McCarty et al, 1998). Biofouling was controlled using pulsed addition of the substrate and H<sub>2</sub>O<sub>2</sub>. The in situ bioremediation system treated trichloroethylene by stimulating toluene-degrading bacteria through the injection of toluene and oxygen. Groundwater was circulated between two contaminated aquifers through two treatment wells placed 10 m apart. The first treatment well (T1) withdrew groundwater from the upper aquifer and discharged it into the lower aquifer, while the second well (T2) performed the reverse function at a flow rate of 38 L/min.

The pumping heads within the system typically increased over time as a result of biomass buildup near the treatment wells, within the aquifer itself. This biofouling was controlled through two different strategies: pulsing the primary substrate (toluene) to optimize its distribution within the aquifer prior to biodegradation, and adding H<sub>2</sub>O<sub>2</sub>, a biocide. A larger amount of H<sub>2</sub>O<sub>2</sub> was added at T1 (71 mg/L) and appeared more effective than the 47 mg/L added at T2 for reducing the total pumping head. Unfortunately the addition of H<sub>2</sub>O<sub>2</sub> decreased the amount of toluene removed, and toluene removal ceased entirely at higher H<sub>2</sub>O<sub>2</sub> (256 mg/L) concentrations. Overall, H<sub>2</sub>O<sub>2</sub> addition appears beneficial for the prevention of biofouling within the system; however, its main disadvantage is its expense. Also, H<sub>2</sub>O<sub>2</sub> can adversely affect the treatment process if used at too high a concentration.

#### **3.2.3.5 Impressed Current Systems**

An applied electrical field using an impressed current system was used as a means of reducing biofilm growth in a water well in Saskatchewan, Canada (Globa and Rohde, 2003). The well was 305 mm in diameter and 21 m deep, with 9.45 m of a stainless steel screen. A pumping test performed after installation in 1995 in the sand and gravel aquifer showed an original specific capacity of 20 imperial gallons per minute (igpm)/ft. In January 2003, the wells specific capacity was measured to be 17.2 igpm/ft, and microbiological testing indicated the presence of slime-forming bacteria.

To mitigate biofouling, four mixed metal oxide (MMO) anodes were installed within the porous media surrounding the stainless steel well. A cathodic protection rectifier was then used to energize the anodes and establish an electrical field in the biofouled porous media. Each anode string was centered at the mid depth of the well screen and spaced 90° apart and 1.5 m away from the screen. The initial voltage level was 20 V, which was increased to 60 V over a 3-month period. During this period the specific capacity stayed relatively constant, indicating that the applied electrical field was successful in maintaining the existing specific capacity (Globo and Rohde, 2003). Because of the absence of a control well, it is difficult to assess whether the impressed current system was superior to having no biofouling control.

### **3.2.3.6 Ultrasonic Tube Resonator**

An ultrasonic tube resonator (UTR) developed by Telsonic Inc. was installed by GeoSyntec in an injection well to curb biofouling in a bioremediation pilot system at a site in Nevada. Ultrasonic technology is commonly used by industry to clean particulate and biological matter from industrial materials and surfaces through the process of cavitation. The UTR generates high-frequency sound waves that expand and contract at a frequency beyond the range of human hearing (greater than 18 kHz). When the sound waves are created in a liquid medium (e.g., groundwater in a vessel or well screen), their expansion phase creates tiny bubbles (cavities). During the contraction phase, these cavities implode, momentarily superheating a microscopic area around them. By imploding near a surface (i.e., the well screen), the energy associated with the superheating will release any particulate matter from the surface (Vaccari, 1999). Because of the small size of the superheated region, the water will heat extremely slowly, and the rise in temperature is not recordable.

During the early stages of pilot test operation, the 3-ft long UTR was periodically (i.e., daily to weekly) raised and lowered over the entire length of the well screen to prevent biofouling. Applications of the UTR occurred variably before, during, and after electron donor injections. Unfortunately, the UTR device, while simple in concept, was found to be cumbersome to deploy and unreliable, suffering several breakdowns related to extended operating time or weather conditions. Therefore, further use of the UTR was abandoned. The UTR was able to maintain the well screen by keeping it clean, but injection pressures continued to rise. Subsequently, samples of the injection well filter pack were collected and it was determined that the plugging around the well was due predominantly to clays and silts held together with polysaccharide slime, with more clay than biomass. This observation suggests that filtration, in addition to physical or chemical biofouling control agents, may be beneficial in preventing fines from plugging the screen, filter pack, and aquifer matrix.

### **3.2.3.7 Summary**

While these studies provide some evidence that biofouling controls have been employed successfully, the reports are limited and often not independently verified. Operational and well design approaches may also minimize fouling, although the results of such approaches are not well documented. Operational procedures can include pulsed nutrient addition, which reduces the amount of time that nutrients are available to promote fouling within the well screen. Well design approaches, such as sizing the well screen and filter pack so as to minimize fouling, have

also been considered, but there is little documented evidence that specific well designs have successfully curtailed well fouling issues.

It is evident that further research is necessary to determine appropriate biofouling controls for bioremediation systems. The advent and validation of safe and reliable preventative biofouling controls is of high value to the continuing use and success of EISB applications. The following sections review and evaluate a wide variety of biofouling controls that may be relevant to groundwater EISB applications.

## **4.0 REVIEW AND INITIAL EVALUATION OF BIOFOULING CONTROLS**

Although biofouling controls have not been used widely for EISB applications, there are significant operational and cost benefits to employing suitable biofouling controls that can be applied to avoid or minimize well rehabilitation. This section explores characteristics of the ideal biofouling control for EISB and discusses general classes of biofouling controls. In Section 5, several of the most promising biofouling controls are discussed in further detail.

### **4.1 CHARACTERISTICS OF IDEAL BIOFOULING CONTROLS FOR EISB**

An ideal biofouling control for EISB applications possesses the following characteristics:

- Exerts the requisite antifouling activity within the well and filter pack, while not persisting so long that the desired contaminant biodegradation reactions are inhibited within the aquifer
- Relatively low dose and/or frequency of application
- Low cost
- Safe and easy to handle, store, and apply
- No adverse impacts on groundwater geochemistry, such as introduction or creation of regulated compounds, mobilization of metals, precipitation of metals causing clogging, undue gas production, significant persisting pH changes
- Addition is readily automated to reduce labor requirements and cost
- Achieves ready regulatory acceptance for addition to aquifer.

Based on these characteristics, biofouling controls utilizing automated hydraulic pressure, mechanical force, heat, or some form of energetic discharge (e.g., UV, electrohydraulic discharge) are particularly attractive since they minimize geochemical impacts and avoid the need for chemical addition. Unfortunately, few technologies/measures of this nature have been developed for preventative control for EISB applications, and as such, chemical biocides are still largely employed. Most chemical additives cause at least some alteration of the geochemical environment. The extent and persistence of these impacts is the key to assessing suitability of a given biocide at a given site.

### **4.2 IDENTIFICATION OF BIOFOULING CONTROLS**

An initial literature survey identified approximately 30 biofouling controls that have been used in well rehabilitation and in a variety of drinking water and industrial processes. These biofouling controls can be classified as oxidizing biocides, nonoxidizing biocides, dispersing agents, physical, and other methods. Table 3 lists the identified biofouling controls by class and provides a description of the biofouling control and its advantages, disadvantages, and uncertainties for use in EISB applications. The major classes of biofouling controls are described further below.

**Table 3. Identification of Available and Emerging Biofouling Controls.**

Biofouling Control	Process Description	Advantages	Disadvantages	Uncertainties	
<b>Oxidizing Biocides</b>	Chlorine	Addition of chlorine gas to the process stream to form a disinfecting residual concentration	Wide spectrum of activity; commodity chemical and equipment used in drinking water treatment; effective disinfectant	Readily consumed by reactions with constituents of biofilm matrix; chlorine gas may generate toxic byproducts (trihalomethanes); performance impacted by pH (decreases at higher pH); handling concerns with chlorine gas; increases redox	Addition to groundwater may be regulated; no guidance on dosing levels, long-term performance, or geochemical impacts
	Sodium hypochlorite	Addition of aqueous sodium hypochlorite to form a disinfecting residual concentration	Wide spectrum of activity; commodity chemical; effective disinfectant	Readily consumed by reactions with constituents of biofilm matrix; generation of trihalomethanes; performance impacted by pH ; increases redox	Addition to groundwater may be regulated; no guidance on dosing levels, long-term performance, or geochemical impacts
	Chlorine dioxide	Addition of chlorine dioxide gas to form a short-lived disinfecting residual concentration	Wide spectrum of activity; commodity chemicals and equipment used in drinking water treatment; highly reactive and effective as biofouling control in groundwater with limited migration from point of introduction; does not form trihalomethane compounds like other chlorination processes	Increases redox; handling considerations with some chlorine dioxide processes that use chlorine gas mixtures; chlorine dioxide nonspecific so high total organic carbon will consume ClO <sub>2</sub>	Effectively used for biofouling control in past; minimal uncertainty
	Bromination	Similar to chlorination; addition of bromine to the process stream to form a disinfecting residual concentration	Stronger disinfectant than chlorine at alkaline pH; wide spectrum of activity	Readily consumed by reactions with constituents of biofilm matrix; not as widely used as chlorine; requires high concentrations; toxic disinfection byproducts may be formed	Not demonstrated in groundwater applications; addition to groundwater may be regulated; no guidance on dosing levels, long-term performance, or geochemical impacts

**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control		Process Description	Advantages	Disadvantages	Uncertainties
<b>Oxidizing Biocides (continued)</b>	Iodine	Similar to chlorination; addition of iodine to the process stream to form a disinfecting residual concentration of iodine/hypiodous acid	Comparable reactivity to chlorine; widely employed in the food industry and as a disinfecting agent for swimming pools and cooling towers	Ineffective against spore-forms of microorganisms; implicated in thyroid toxicity; can produce halogenated disinfection products; iodine vapor is toxic	Limited use in groundwater applications; addition to groundwater may be regulated; no guidance on dosing levels, long-term performance, or geochemical impacts
	Ozone	Addition of ozone to the process stream, which acts as a disinfectant	Wide spectrum of activity; highly reactive with limited migration from point of introduction; hydrolyzes exopolysaccharides	Readily consumed by reactions with constituents of biofilm matrix or total organic carbon; potential to form bromate byproducts; highly corrosive; requires ozone generators, which are available in a limited range of sizes; decomposes to O <sub>2</sub> , resulting in aerobic/ oxidizing conditions	Limited use in groundwater applications; no guidance on dosing levels, long-term performance, or geochemical impacts
	Hydrogen peroxide	Addition of hydrogen peroxide to the process stream to form a disinfecting residual concentration	Readily applied; commodity chemical and equipment; highly reactive in groundwater with limited migration from point of introduction; degrades exopolysaccharides	Highly reactive with common mineral types, potentially resulting in heat/vapor generation; decomposes to O <sub>2</sub> , resulting in aerobic/oxidizing conditions	Limited use in groundwater applications; no guidance on dosing levels, long-term performance, or geochemical impacts
	Peracetic acid	Addition of an acetic acid/hydrogen peroxide mixture to the process stream to form a disinfecting residual concentration	Wide spectrum of activity at low concentration; penetrates biofilms; non-toxic	Corrosive and unstable; handling a potential concern; decomposes to acetic acid, an electron donor readily used by methanogens	Limited use in groundwater applications; no guidance on dosing levels, long-term performance, or geochemical impacts; vendors may be difficult to locate

**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control	Process Description	Advantages	Disadvantages	Uncertainties	
<b>Non-Oxidizing Biocides</b>	Alcohols	Pulsed addition of an alcohol (used as electron donor) at a biocidal concentration into the process stream	Readily available commodity chemical; easy to apply; may be utilized as electron donor to support biodegradation	Limited effectiveness; flammability imposes handling costs; requires a high concentration; high taxation rates on purchase of some alcohols (especially EtOH); may encourage biofouling	Addition to groundwater may be regulated; no guidance on dosing levels or long-term performance
	Glutaraldehyde	Addition of glutaraldehyde to the process stream to form a disinfecting residual concentration	Effective in low concentrations; inexpensive, noncorrosive; degrades to formic acid, which may be used as an electron donor to support biodegradation; does not raise redox making it favorable for anaerobic processes; biodegradable under aerobic conditions; test kits available	Limited penetration of biofilm matrix; at high concentrations may not be compatible with PVC; registered pesticide that requires SLNs permit for use in groundwater	No performance data for groundwater applications; addition to groundwater may be prohibited; may adversely impact degrading microorganisms; no guidance on dosing levels or long-term performance
	Tetrakis(hydroxy-methyl) phosphonium sulfate (THPS)	Addition of THPS to the process stream to form a disinfecting residual concentration	Highly effective against sulfate reducing bacteria; wide spectrum of activity; safe to handle; non-corrosive; penetrates biofilm; does not raise redox, making it favorable for anaerobic processes; biodegrades under aerobic and anaerobic conditions; test kits available to monitor concentrations	No data on long-term fate in groundwater, including impacts on dechlorinating microorganisms; no impact on biofilm matrix; contains trace levels of formaldehyde; registered pesticide that requires SLNs permit	Limited use in remediation applications; addition to groundwater may be prohibited; may adversely impact degrading microorganisms; no guidance on dosing levels, or long-term performance
<b>Chelating and Dispersing Agents</b>	Citric acid	Addition of citric acid to the process stream to destabilize and disperse biofilm	Safe to handle; destabilizes biofilm matrix; commodity chemical; can act as an electron donor to support biodegradation	Lowers pH of injected groundwater; may increase biofouling; may mobilize metals	Limited use in remediation applications; no guidance on dosing levels, long-term performance, or geochemical impacts

**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control	Process Description	Advantages	Disadvantages	Uncertainties	
<b>Chelating and Dispersing Agents (continued)</b>	Polymaleic acid	Addition of polymaleic acid to the process stream to destabilize and disperse biofilm	Safe to handle; destabilizes biofilm matrix; commercially available as NW310 (Johnson Screens), widely used in water well rehabilitation; can act as an electron donor to support biodegradation	Lowers pH of injected groundwater; may mobilize metals	No guidance on dosing levels, long-term performance or geochemical impacts
	Glycolic (hydroxyacetic) acid	Addition of hydroxyacetic acid to the process stream to destabilize and disperse biofilm	Safe to handle; destabilizes biofilm matrix; exerts a bactericidal effect; widely used in water well rehabilitation; verbal reports of successful applications as a biofouling control agent in groundwater application	Lowers pH of injected groundwater; may mobilize metals	No information on long-term performance or geochemical impacts
	Surfactants	Addition of surfactant to the process stream to destabilize and disperse biofilm	Penetrates and disperses biofilm matrix; permits more effective application of biocides; widely used in water well rehabilitation; surfactants likely to biodegrade	Likely requires high concentrations; high chemical cost; may be more effective for rehabilitation rather than prevention	No guidance on dosing levels, long-term performance, or geochemical impacts
	Polyphosphates	Addition of concentrated solution into the injection point to destabilize and disperse biofilm	Destabilizes biofilm matrix and disperses clays; widely used in water well rehabilitation	Addition into phosphate-limited nutrient condition may promote a rapid increase in microbial growth	No guidance on dosing levels, long-term performance, or geochemical impacts
	Enzymes	Addition of specific enzymes (e.g., pectinase) into the process stream to break down pectin or other components in the biofilm matrix	Targets biofilm matrix; emerging laboratory studies evaluating the use of pectinase with promising results at high concentrations	Potentially cost prohibitive; no documented field applications; enzyme may have low activity at pH and temperature of groundwater	Not a commercial technology; unknown effects on biofilm formation and beneficial microorganisms (contaminant degraders)



**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control		Process Description	Advantages	Disadvantages	Uncertainties
<b>Physical</b>	Brushing	Application of a mechanical tool to physically remove particles from the well screen	Easy to apply; highly effective at removing deposits from well screen; minimal geochemical impacts	Limited removal of biofilm from filter pack; may require specialized equipment and removal of debris	Not typically applied to injection wells during operation; would likely require frequent application
	Surging/swabbing	Use of downhole tools to generate fluid shear stress and enhance biofilm detachment from the well screen/filter pack and debris mobilization into the well	Easy to apply; highly effective at removing deposits from well screen and filter pack	May require specialized equipment and removal of debris	Not typically applied to injection wells during operation; would likely require frequent application
	Jetting	Injection of water or other fluids at high rate into the injection well to physically remove biofouling from well screen/filter pack	Easy to apply; highly effective at removing deposits from well screen, some removal from filter pack	May require specialized equipment and removal of debris	Not typically applied to injection wells during operation; would likely require frequent application
	Carbon dioxide (Aqua Gard™)	Injection of liquid and gaseous carbon dioxide into the formation, followed by extraction of the gas and dislodged particles	Effective on bacteria and biofilm matrix; creates physical, chemical, and thermal impacts; non-toxic byproducts	Currently there are few vendors for this technology; potential for damage to well screen/casing; formation of carbonic acid may result in pH drop	Limited case studies and vendors available
<b>Other</b>	Ultrasound	Use of ultrasonic energy generated in the well casing by a downhole generator to remove scale and destabilize biofilm; creates pressure waves and cavitation	Highly effective at removing scale under static conditions; chemical not required; easy to apply	Performance likely to be improved when applied in conjunction with a biocide; limited industrial application; high cost for individual transducer units; high temperature may impact transducer reliability; may require shutdown of system prior to application; limited range, needs to be moved along the well screen	Limited testing has shown ultrasound to be effective at removing biofilm from the well screen but not the filter pack.

**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control	Process Description	Advantages	Disadvantages	Uncertainties	
<b>Other (continued)</b>	Plasma/electro-hydraulic discharge	Use of electrohydraulic energy to sterilize process water, degrade soluble electron donors, and destabilize biofilms; creates pressure waves, cavitation, and oxidant formation; applied to either process water or within the well	Potentially good penetration into the filter pack; chemicals not required, easy to apply	Performance likely to be improved when applied in conjunction with a biocide; limited industrial application; high cost for individual emitter units; environmental conditions may impact emitter reliability	Few commercial providers of this technology
	Acoustic	Use of acoustic energy to disinfect water; used in marine applications to treat fouling in heat exchangers and piping	No chemicals are added so no toxic or geochemical impacts	May need to move device up and down well screen; cost may be prohibitive	Equipment not commercially available; no information on use in groundwater wells
	Bacteria/bacteriophage	Addition of microorganisms or bacteriophages (viruses) into the process stream that feed on or destroy bacteria in the biofilm	Potentially low impacts on groundwater geochemistry; targets both bacteria and biofilm matrix	Possibly highly specific to a limited number of bacteria; unproven in any application; bacteriophage will have no impact on exopolysaccharides or mineral scale; phage addition may promote fouling	Not a commercial technology; possible regulatory constraints on the addition of bacteriophages
	Thermal pasteurization	Application of heat into the injection point and the surrounding formation to create a biocidal effect	Biocidal effect in the well screen and surrounding filter pack; used in conjunction with biocides & surfactants, heat improves chemical reactivity; commercially applied for water supply well rehabilitation, usually in conjunction with other chemicals	High energy costs; may stimulate microbial growth if improperly applied; potential to form carbonate scale	Limited guidance on heating requirements; may stimulate growth of thermophilic microorganisms

**Table 3. Identification of Available and Emerging Biofouling Controls (continued).**

Biofouling Control	Process Description	Advantages	Disadvantages	Uncertainties	
<b>Other (continued)</b>	Muriatic acid (HCl)	Addition of concentrated acid into the injection point	Highly effective in removing scale; widely used in water well rehabilitation	Not effective against iron fouling; produces toxic fumes, requires careful handling; product potentially contains trace impurities; lowers pH significantly	No guidance on dosing levels, long-term performance, or geochemical impacts
	Sulfamic acid (H <sub>3</sub> NO <sub>3</sub> S)	Addition of concentrated acid into the injection point; widely used in water well rehabilitation	Safer to handle relative to muriatic acid	Not effective against iron or manganese scaling; requires careful handling; potentially contains trace impurities; lowers pH significantly	No guidance on dosing levels, long-term performance, or geochemical impacts
	Ultraviolet (UV) disinfection with filtration	UV system transfers electromagnetic energy to the injected stream, inactivating microorganisms DNA and RNA, destroying their ability to reproduce; filtration to remove solids	Does not involve the addition of chemicals; should have minimal impact on water quality if applied on injected stream; at low doses should not alter the contaminants of interest; has been used for groundwater disinfection on water wells	No impact on existing or established biofilms; injection well may require presterilization first	No guidance for use as a biofouling control agent
	Impressed current system	Anodes electrically coupled to well screen, establishing an electrical field in the porous media surround the well screen; electrical current causes biofilm to shrink; mechanism not well understood	Does not involve the addition of chemicals; should have minimal impact on water quality	Cost involved with installation of electrodes; has shown limited effectiveness for treating biofouling in water wells	Has not been tested in groundwater remediation applications
	Filtration	Removal of microorganisms in influent stream	No geochemical impacts	High maintenance costs and high pressure drops possible; sterilization of injection well still required	No guidance for use as a biofouling control agent

**Oxidizing biocides** include chlorine, chlorine dioxide, sodium hypochlorite (NaOCl), bromine, iodine compounds, ozone, H<sub>2</sub>O<sub>2</sub>, and peracetic acid. Oxidizing biocides have been used extensively in water treatment applications and have demonstrated disinfection capabilities, making them advantageous for use. The disadvantage of oxidizing biocides, as a group, is that they increase the redox potential of the aquifer, which can cause mineral precipitation and can lower the efficiency of an anaerobic bioremediation process requiring reduced conditions. Most oxidizing biocides also require special handling precautions.

**Nonoxidizing biocides** include compounds such as alcohols, glutaraldehyde, and tetrakis (hydroxy-methyl) phosphonium sulfate. The advantage of this class of chemicals is that they do not significantly impact the redox potential of the system, and have proven biocidal capabilities in other applications. These biocides are also typically biodegradable. The disadvantages of using alcohols are their cost and flammability. Glutaraldehyde, and tetrakis (hydroxy-methyl) phosphonium sulfate have low toxicity relative to other biocides but have not been used extensively in groundwater applications. Consequently, their use may meet with some resistance from regulatory agencies and may require special permits.

**Dispersing agents** act primarily to break down and disperse the biofilm matrix rather than to kill or inactivate the bacteria within the biofilm. Dispersing agents include citric acid, polymaleic acid, glycolic acid, surfactants, polyphosphates, and enzymes. The organic acids (citric, polymaleic, and glycolic acids) disperse biofilms primarily by acting as chelating agents and extracting calcium or magnesium from the biofilm. Surfactants and polyphosphates act primarily to disperse the biofilm by penetrating it. Surfactants and polyphosphates are widely used in well rehabilitation to improve the contact between disinfectants and the biofilm, but they may act to encourage biofouling because they are readily biodegraded. Enzymes, such as pectinase, act to degrade pectin in the exopolysaccharides within the biofilm matrix. The advantages of dispersing agents are that, with the exception of enzymes, they have been used successfully in well rehabilitation applications and have therefore gained regulatory acceptance. Their disadvantages include decreasing the pH (in the case of acid addition) and possibly promoting biofouling through the addition of a readily degradable compound (such as a surfactant or polyphosphate compound) or by breaking down the biofilm matrix, which is itself biodegradable.

**Physical** control measures include brushing, surging or swabbing, jetting, and carbon dioxide (Aqua Gard<sup>TM</sup>). The advantages of physical biofilm removal approaches are that they are non-toxic, do not impact the environmental conditions of the aquifer beyond temporarily increasing the turbidity, and are more acceptable to the regulatory community. The disadvantage is that biofouling returns relatively rapidly, and brushing does not remove bacterial growth in the filter pack.

The “**Other**” category captures a wide array of biofouling controls, some of which are emerging in nature, including ultrasound, plasma/electrohydraulic discharge, bacteriophages, thermal pasteurization, acid addition, impressed current systems, and wellhead pretreatment technologies (such as UV irradiation and filtration). The advantage of ultrasound, plasma/electrohydraulic discharge, and bacteriophages is that they should all have minimal impact on geochemistry. The downside is that many are not commercially available or may be prohibitively expensive or

impractical to apply in a preventative manner. The acids include muriatic and sulfamic acid. Advantages of acids include their widespread use in well rehabilitation applications and their effectiveness in removing inorganic scale. Disadvantages include safety and cost considerations associated with handling strong acids (e.g., muriatic acid) and the presence of trace impurities in both acids. Thermal pasteurization has the advantage that it does not involve the addition of chemicals. Its disadvantages include the possibility of increasing biofouling at some distance away from the injection well and decreasing the solubility of carbonates in the aquifer. Impressed current systems have not been shown to definitively prevent biofouling, but have the advantage of minimal geochemical impacts. Conventional well-head pretreatment may inactivate or remove bacteria and solids from the injected groundwater/fluids, reducing bacterial and solids loading to the injection wells and hence reducing the rate of fouling. However, biofouling could still occur in the filter pack and surrounding aquifer, constraining injection well performance.

### **4.3 INITIAL EVALUATION AND SCORING OF BIOFOULING CONTROLS**

To evaluate and compare the suitability of the individual EISB biofouling controls identified in Table 3, a scoring system was established based on eight technical, financial, and regulatory criteria. Table 4 presents the results of the evaluation and scoring process. Biofouling controls were scored relative to each other in each category, with the objective of selecting the most promising control in each category. From this process, a subset of biofouling controls (primary and secondary) was selected for detailed evaluation (see Section 5) and field testing as part of subsequent activities for ESTCP Project ER-0429. The evaluation and scoring criteria are defined below. Scoring for each criteria was based on a five-point scale (40-point maximum total score), with one point being lowest suitability and five being highest suitability for widespread acceptance and use.

**EISB compatibility** refers to the ability of the control to be used for a wide range of EISB processes, such as reductive dechlorination, anaerobic oxidation, aerobic oxidation, and/or co-oxidation. Many of the identified biofouling controls have the potential to influence groundwater redox potential, making the control unsuitable for use in specific geochemical environments and across the aforementioned range of degradation mechanisms potentially employed during EISB applications. For example, the use of oxidizing biocides is likely to disrupt anaerobic microbial activity and is unlikely to be coupled to reductive dechlorination processes, whereas physical measures such as ultrasound are independent of redox potential and geochemistry and can be widely used, independent of degradation mechanism.

**Commercial availability** reflects the degree to which a biofouling control is widely (geographically) available off-the-shelf versus in research and development stages. Several controls have been demonstrated to be effective at laboratory scale but have not yet been evaluated or validated in appropriate scale field demonstrations.

**Implementability** reflects the level of complexity for implementation of the biofouling control. For example, the use of nonoxidizing biocides requires very little infrastructure (mainly just an ex situ storage tank and metering pump), whereas many of the controls in the Other category require complicated down-well infrastructure that may not be suitable for automation.

**Table 4. Evaluation and Scoring of Biofouling Controls.**

Biofouling Controls		Evaluation Criteria / Scoring								Total Score (maximum of 40)
		Proven/Likely Effectiveness	EISB Compatibility	Commercial Availability	Implementability	Handling and Safety	Regulatory Acceptance	Water Quality Impacts	Cost	
<b>Oxidizing Biocides</b>	Chlorine	4	3	5	4	1	3	2	3	25
	Sodium hypochlorite	4	3	5	5	4	3	2	4	<b>30*</b>
	Chlorine dioxide	5	3	5	4	3	4	5	3	<b>32</b>
	Bromination	3	3	3	4	2	3	2	4	24
	Iodine	3	3	3	3	2	3	3	4	24
	Ozone	4	3	4	3	2	4	3	2	25
	Hydrogen peroxide	3	3	5	4	3	4	3	3	28
Peracetic acid	3	3	3	4	3	3	3	3	25	
<b>Nonoxidizing Biocides</b>	Alcohols	2	2	4	3	2	4	3	2	22
	Glutaraldehyde	3	4	4	4	3	1	4	4	27
	THPS	4	4	4	4	4	2	4	4	<b>30</b>
<b>Chelating and Dispersing Agents</b>	Citric Acid (HOC(COOH)(CH <sub>2</sub> COOH) <sub>2</sub> )	3	3	5	5	4	4	3	4	31
	Polymaleic acid	3	3	5	5	4	4	3	4	31
	Hydroxyacetic acid	4	3	5	5	4	4	3	4	<b>32</b>
	Surfactants	2	4	4	4	5	4	3	4	30
	Polyphosphates	2	4	4	4	5	4	2	4	29
	Enzymes	1	4	2	3	5	3	3	1	22
<b>Physical Controls</b>	Brushing/swabbing/surging	3	4	4	3	4	5	5	2	30
	Jetting	3	4	4	3	4	5	5	2	30
	Carbon dioxide (Aqua Gard™)	4	5	4	3	3	5	4	3	<b>31</b>
<b>Other</b>	Ultrasound	3	5	3	2	4	5	5	3	30
	Plasma/electrohydraulic discharge	2	5	1	2	4	4	5	?	23
	Acoustic	2	5	1	2	4	4	5	?	23
	Bacteria/bacteriophage	1	2	1	2	4	1	3	?	14
	Thermal pasteurization	4	4	4	4	4	4	4	3	<b>31</b>
	Muriatic acid (HCl)	2	3	5	3	1	3	2	3	22
	Sulfamic acid (H <sub>2</sub> NO <sub>3</sub> S)	2	3	5	3	2	3	2	3	23
	Ultraviolet radiation with filtration	2	4	5	4	4	5	5	2	31
	Filtration	1	5	5	2	5	5	5	3	31
Impressed current system	2	5	4	3	4	5	5	?	28	

Notes:

Scoring: 1 Lowest (Worst) vs 5 Highest (Best)

\*Biofouling Control Technologies highlighted in bold are primary biofouling control candidates for the field demonstration

**Proven/likely effectiveness** reflects the maturity and degree of performance validation data available for a given biofouling control. Controls such as chlorine dioxide have proven successful at several sites at reasonable scale (10 to 150 gpm systems) and durations (up to 300 days). Acoustic controls have yet to be tested in field EISB applications, but laboratory data suggests the approach may be promising.

**Handling and safety** are primary concerns for all remedial applications. The use of many of the physical controls carries little risk with regard to handling of the equipment and materials, or to the safety of practitioners, whereas the use of many of the biocides (particularly the oxidizing biocides) has significant handling issues related to human safety.

**Regulatory acceptance** reflects the known or anticipated response to proposed use of a given biofouling control for groundwater EISB applications. Some of the chemical biocides are quite recalcitrant, and therefore, regulatory authorities may be reluctant or unwilling to permit their addition to groundwater. By comparison, regulators are unlikely to take issue with the use of many of the physical controls.

**Water quality impacts** reflects the degree to which a given biofouling control is known or anticipated to cause a new or adverse impact to groundwater quality. For example, the addition of halogenated oxidizing biocides may generate trihalomethanes, which are regulated compounds, whereas acids may alter groundwater pH and mobilize dissolved metals (e.g., arsenic, manganese, iron) from aquifer materials.

The cost metric considers the capital cost of equipment required to instrument a nutrient delivery well, O&M costs (labor, chemicals, power, health and safety) related to the control. The dose, persistence, and frequency of application for most chemical biocides will significantly dictate O&M costs.

A more detailed cost comparison can be found in Table 5, where only biofouling controls that were likely to be effective (i.e., that scored higher than 2 in the Proven/Likely effectiveness category) were evaluated. Capital costs and anticipated annual O&M costs are provided. Capital costs were categorized as low, medium, or high, which corresponded to <\$5K, \$5-\$10K, and >\$15K. Generally, these costs are for a 5-50 gpm system; however, the cost for some biofouling controls were estimated per well (as in the case of the ultrasound tube resonator and the Aqua Gard™ technology). Anticipated annual operating and maintenance costs per well were also estimated and categorized as low (<\$3K), medium (\$3-10K), and high (>\$10K). Annual O&M costs are difficult to determine in advance of the demonstration because they are highly dependent on the effectiveness of the biofouling control and the frequency of application. The annual O&M costs will be refined after the demonstration for the biofouling controls tested in the field.

Nonoxidizing biofouling and chelating and dispersing agents had low capital and anticipated annual operating costs because their application requires only a feed tank, dosing pump, and controls, biofouling controls that are easily automated. Oxidizing biocides are, on average, more expensive as some require expensive generators and safety features, but they are generally easy to automatically apply. Physical methods can be more expensive than nonoxidizing biocides,

since they are more difficult to automate. Lastly, ultrasound and thermal pasteurization are expected to have low annual O&M costs per well but have higher capital costs.

**Table 5. Cost Comparison of Biofouling Controls.**

<i>Biofouling Controls</i>		<i>Anticipated Capital Cost for 5-50 gpm</i>	<i>Anticipated Annual O&amp;M per Well</i>
<b>Oxidizing biocides</b>	Chlorine	med	low
	Sodium hypochlorite	low	low
	Chlorine dioxide	med	low
	Bromination	low	low
	Iodine	low	low
	Ozone	med-high	med
	Hydrogen peroxide (H <sub>2</sub> O <sub>2</sub> )	low	low-med
<b>Nonoxidizing biocides</b>	Peracetic acid	low	med
	Glutaraldehyde	low	low
<b>Chelating and</b>	THPS	low	low
	Citric acid (HOC(COOH)(CH <sub>2</sub> COOH) <sub>2</sub> )	low	low
<b>Dispersing agents</b>	Polymaleic acid	low	low
	Glycolic acid	low	low
<b>Physical</b>	Brushing/swabbing/surgin	low	high
	Jetting	low	med-high
	Carbon dioxide (Aqua Gard™)	low-med*	med
<b>Other</b>	Ultrasound	med-high*	low
	Thermal pasteurization	med	low

Notes:

	Capital	O&M
low	<\$5K	<\$3
med	\$5-15K	\$3-10K
high	>\$15K	>\$10K

\* These capital costs are per well, whereas the other capital costs are per system.



*This page left blank intentionally.*

## 5.0 DETAILED EVALUATION OF PROMISING BIOFOULING CONTROL OPTIONS

As indicated in Section 4.3, the various biofouling controls were evaluated and scored in Table 4 to select a subset of biofouling controls for detailed evaluation and field testing as part of ESTCP Project ER-0429. Based on the design of the field demonstration/validation project (to be presented in a separate ESTCP Demonstration Plan), the performance of five biofouling controls will be concurrently evaluated. Given that some biofouling controls may fail in a relatively short time frame, both primary and secondary controls employing similar infrastructure were identified for field testing, and attempts were made to select controls from each of the main classes of controls to diversify testing. The demonstration will begin using the primary controls. If a primary control fails within 60 days, the delivery well will be retrofitted for use of the secondary control, to the extent possible, for the remainder of the demonstration.

Table 6 identifies the primary and secondary controls identified for use in the field trial. The highest scoring control in Table 4 in each category was selected as the primary control. Although nonoxidizing biocides such as THPS and glutaraldehyde appear promising, their status as pesticides and the time associated with acquiring an SLN permit precluded their selection for the demonstration. Instead, a second oxidizing biocide was chosen, given their proven disinfection capability and common use. The primary controls identified for the biofouling demonstration include:

- *Oxidizing Biocide #1*: chlorine dioxide
- *Oxidizing Biocide #2*: sodium hypochlorite
- *Chelating or Dispersing Agent*: glycolic acid (LBA)
- *Physical*: carbon dioxide (Aqua Gard™)
- *Other*: thermal pasteurization.

The following sections provide detailed descriptions of these primary biofouling controls, including the mode of action, typical applications, potential impacts to water quality, and anticipated implementation issues.

### 5.1 CHLORINE DIOXIDE

#### 5.1.1 Mode of Action

Chlorine dioxide is a strong oxidant and disinfectant. It is an effective biocide at concentrations as low as 0.1 ppm and over a wide pH range. Its disinfection mechanism is not well understood; however, it is believed that chlorine dioxide likely penetrates the bacteria cell wall and reacts with vital amino acids in the cytoplasm of the cell to kill the organism (Noss et al, 1983). Because chlorine dioxide is more soluble and has greater oxidative capacity than chlorine (five electrons to two), chlorine dioxide can be 10 to 50 times more effective than chlorine.

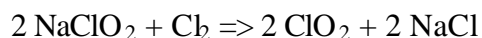
**Table 6. Proposed Primary and Secondary Biofouling Controls for Field Demonstration/Validation.**

Primary Biofouling Control	Secondary Biofouling Control (Time And Budget Permitting)
Chlorine dioxide (gas) ClO <sub>2</sub>	Chlorine dioxide (aqueous)
Sodium hypochlorite	Hydrogen peroxide
Glycolic acid	Polymaleic acid
Carbon dioxide (Aqua Gard™)	Citric acid
Thermal pasteurization	Ultrasound

### 5.1.2 Generation of Chlorine Dioxide

As chlorine dioxide is pressure-sensitive and will decompose if it is compressed for storage or shipping (Gates, 1998), it must be manufactured on site.

There are several different processes for generating ClO<sub>2</sub>, many of which are discussed in Environmental Protection Agency (EPA) guidance on alternative disinfectants (EPA, 1999). A summary of these processes is provided in Table 7. In the chlorine dioxide generator (CDG) process (Bethlehem, Pennsylvania), which has been used with success in several EISB applications, a blend of compressed chlorine gas in nitrogen is passed through a reactor cartridge containing specially-processed NaClO<sub>2</sub>. The chlorine reacts with the NaClO<sub>2</sub> in the following reaction:



For each mole of chlorine, the process generates two moles of ClO<sub>2</sub>. As long as the chlorine feed gas concentration never exceeds 4% in nitrogen, the concentration of chlorine dioxide cannot enter the explosive range (>10%). The chlorine dioxide is dissolved in a water stream and the aqueous solution is applied to the well.

Chlorine dioxide can also be produced from hydrochloric or hypochlorous acid (HOCl) and NaClO<sub>2</sub> (EPA, 1999) or electrochemically (Pureline, Lake Forest, California) with NaClO<sub>2</sub> as the only feed chemical (Gates, 1998). While these processes have been used in industrial applications, their use in EISB applications has not been documented.

### 5.1.3 Where Used

ClO<sub>2</sub> is used extensively as a bleaching agent in the pulp and paper industry. ClO<sub>2</sub> is also used in the food industry for fruit and vegetable washing, flume water disinfection, meat and poultry disinfection, sanitizing food process equipment, and for odor control. In industrial processes, chlorine dioxide is used in cooling systems/towers, ammonia plants, pulp mills (slime control, paper machines), oil fields, scrubbing systems/odor control, textile bleaching, and the electronics industry. ClO<sub>2</sub> is also used to control iron and manganese dissolution and hydrogen sulfide and phenolic compound production (EPA, 1999).

**Table 7. Commercial Chlorine Dioxide Generation Systems.**

Generator Type	Main Reactions	Special Attributes
ACID-CHLORITE: (Direct acid system)	$4\text{HCl} + 5\text{NaClO}_2 \rightarrow 4\text{ClO}_{2(\text{aq})} + \text{ClO}_3^-$ <ul style="list-style-type: none"> <li>• Low pH</li> <li>• <math>\text{ClO}_3^-</math> possible</li> <li>• Slow reaction rates</li> </ul>	<ul style="list-style-type: none"> <li>• Chemical feed pump interlocks required</li> <li>• Production limit ~ 25-30 lb/day</li> <li>• Maximum yield at ~80% efficiency</li> </ul>
AQUEOUS CHLORINE-CHLORITE: (Cl <sub>2</sub> gas ejectors with chemical pumps for liquids or booster pump for ejector water)	$\text{Cl}_2 + \text{H}_2\text{O} \rightleftharpoons [\text{HOCl} / \text{HCl}]$ $[\text{HOCl} / \text{HCl}] + \text{NaClO}_2 \rightarrow \text{ClO}_{2(\text{g})} + \text{H} / \text{OCl}^- + \text{NaOH} + \text{ClO}_3^-$ <ul style="list-style-type: none"> <li>• Low pH</li> <li>• <math>\text{ClO}_3^-</math> possible</li> <li>• Relatively slow reaction rates</li> </ul>	<ul style="list-style-type: none"> <li>• Excess Cl<sub>2</sub> or acid to neutralize NaOH</li> <li>• Production rates limited to ~ 1000 lb/day</li> <li>• High conversion but yield only 80-92%</li> <li>• More corrosive effluent due to low pH (~2.8-3.5) (Three chemical systems pump HCl, hypochlorite, chlorite, and dilution water to reaction chamber.)</li> </ul>
RECYCLED AQUEOUS CHLORINE OR "FRENCH LOOP"™ (Saturated Cl <sub>2</sub> solution via a recycling loop prior to mixing with chlorite solution)	$2\text{HOCl} + 2\text{NaClO}_2 \rightarrow 2\text{ClO}_2 + \text{Cl}_2 + 2\text{NaOH}$ <ul style="list-style-type: none"> <li>• Excess Cl<sub>2</sub> or HCl needed due to NaOH formed</li> </ul>	<ul style="list-style-type: none"> <li>• Concentration of ~3 g/L required for maximum efficiency</li> <li>• Production rate limited to ~ 1,000 lb/day</li> <li>• Yield of 92-98% with ~10% excess Cl<sub>2</sub> reported; highly corrosive to pumps; draw-down calibration needed; maturation tank required after mixing</li> </ul>
GASEOUS CHLORINE-CHLORITE (Gaseous Cl <sub>2</sub> and 25% solution of sodium chlorite; pulled by ejector into the reaction column)	$\text{Cl}_{2(\text{g})} + \text{NaClO}_{2(\text{aq})} \rightarrow \text{ClO}_{2(\text{aq})}$ <ul style="list-style-type: none"> <li>• Neutral pH</li> <li>• Rapid reaction</li> <li>• Potential scaling in reactor under vacuum due to hardness of feedstock</li> </ul>	<ul style="list-style-type: none"> <li>• Production rates 5-120,000 lb/day</li> <li>• Ejector-based, with no pumps; motive water is dilution water, near neutral pH effluent</li> <li>• No excess Cl<sub>2</sub>; turndown rated at 5-10X with yield of 95-99%; less than 2% excess Cl<sub>2</sub>; highly calibrated flow meters with minimum line pressure; ~40 psig needed</li> </ul>
GASEOUS CHLORINE-SOLID CHLORITE MATRIX (Humidified Cl <sub>2</sub> gas pulled or pumped through a stable matrix containing solid sodium chlorite)	$\text{Cl}_{2(\text{g})} + \text{NaClO}_{2(\text{s})} \rightarrow \text{ClO}_{2(\text{g})} + \text{NaCl}$ <ul style="list-style-type: none"> <li>• Rapid reaction rate</li> <li>• New technology</li> </ul>	<ul style="list-style-type: none"> <li>• Cl<sub>2</sub> gas diluted with N<sub>2</sub> or filtered air to produce ~8% gaseous ClO<sub>2</sub> stream; infinite turndown possible with &gt;99% yield</li> <li>• Maximum rate to ~1,200 lb/day per column</li> </ul>
ELECTROCHEMICAL (Continuous generation of ClO <sub>2</sub> from 25% chlorite solution recycled through electrolyte cell)	$\text{NaClO}_{2(\text{aq})} \rightarrow \text{ClO}_{2(\text{aq})} + \text{e}^-$ <ul style="list-style-type: none"> <li>• New technology</li> </ul>	<ul style="list-style-type: none"> <li>• Counter-current chilled water stream accepts gaseous ClO<sub>2</sub> from production cell after it diffuses across the gas permeable membrane</li> <li>• Small one-pass system requires precise flow for power requirements (Coulombs law)</li> </ul>
ACID/PEROXIDE/CHLORIDE	$2\text{NaClO}_3 + \text{H}_2\text{O}_2 + \text{H}_2\text{SO}_4 \rightarrow 2\text{ClO}_2 + \text{O}_2 + \text{NaSO}_4 + \text{H}_2\text{O}$	<ul style="list-style-type: none"> <li>• Uses concentrated H<sub>2</sub>O<sub>2</sub> and H<sub>2</sub>SO<sub>4</sub></li> <li>• Downscaled version; foam binding; low pH</li> </ul>

Source: Adapted from Gates, 1998

GeoSyntec has used ClO<sub>2</sub> to control biofouling in electron donor delivery wells at many sites employing EISB (GeoSyntec, 2003; 2004). The CDG used a preblended compressed gas cylinder to supply a pressurized mixture of nitrogen and chlorine gas (96% nitrogen: 4% chlorine). The gas mixture was passed through a cylinder of NaClO<sub>2</sub>, generating 8% ClO<sub>2</sub> in nitrogen. The ClO<sub>2</sub> was piped directly into the recharge water in the injection well daily for 1-hr at a dose of 1 mg/L. ClO<sub>2</sub> was effective in controlling biofouling in the electron donor delivery well for more than 6 mo.

#### **5.1.4 Impacts to Water Quality**

Chlorine dioxide has an advantage over chlorine in that it does not produce chlorinated disinfection by-products, such as chloroform. It does, however, have the potential to produce chlorite (1 mg/L MCL), particularly when natural organic matter is present (Werdehoff and Singer, 1987). A wide variety of bacteria have chlorite dismutase enzymes, which promote rapid decomposition of chlorite. As such, chlorite is not expected to persist or cause adverse impacts to groundwater quality.

Chlorine dioxide is an oxidizing agent and, as such, will increase the oxidation-reduction potential (ORP) of the aquifer. Increased ORP can result in the oxidation of ferrous iron and precipitation of iron oxides, which can result in chemical fouling of the well screen or filter pack. The oxidized conditions may also have a deleterious impact on anaerobic degradation activity in the near vicinity of the delivery wells.

#### **5.1.5 Implementation Issues**

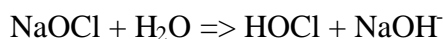
The main implementation issue relates to safety considerations when using chlorine dioxide. Some chlorine dioxide generation processes require the use of a compressed cylinder of chlorine/nitrogen to generate the chlorine dioxide. The use of compressed gas cylinders and the presence of a toxic gas (chlorine) and asphyxiate (nitrogen) require additional safety measures to prevent tampering of the cylinders. Other processes for chlorine dioxide generation processes do not use compressed gas cylinders (see Table 7) and thus may be more desirable for use for safety reasons, provided that they can be shown not to generate trihalomethanes.

As an additional safety issue, chlorine dioxide is explosive at concentrations that exceed 10% by volume in air (EPA, 1999). Therefore, the ClO<sub>2</sub> generation process should be designed to prevent the generation or accumulation of high concentrations of chlorine dioxide. Chlorine dioxide is reactive with natural organic matter and possibly reactive with added organic electron donor, reducing its effectiveness and requiring higher initial doses.

### **5.2 SODIUM HYPOCHLORITE**

#### **5.2.1 Mode of Action**

NaOCl is an oxidizing biocide, having well characterized disinfection properties. When added to water, NaOCl reacts to form HOCl as follows:



The HOCl formed is a relatively weak acid and is very poorly dissociated at pH levels below 6. At higher pH levels, the hypochlorite ion (OCl<sup>-</sup>) is formed (Sawyer et al, 1994). Although the hypochlorite ion has a higher oxidation potential than HOCl, HOCl is much more effective as a disinfectant. Since HOCl has no charge, it is able to penetrate microbial cell walls easier, causing alternations in cellular metabolism and destruction of phospholipids, irreversible enzyme activation, and fatty acid degradation. NaOCl is most effective at a pH between 6.5 and 7.5.

### **5.2.2 Where Used**

NaOCl (better known as bleach) has been used for many years to disinfect drinking water, due to its safety and efficacy. It is commonly used as a bleaching agent or disinfectant in laundries, swimming pools, ponds, drinking water, and other water and wastewater systems (ATSDR, 2002; EPA, 1991). NaOCl has also been used on food and nonfood contact surfaces, and as a postharvest seed or soil treatment on various fruit and vegetable crops (EPA, 1991). NaOCl has been routinely used in well rehabilitation.

### **5.2.3 Impacts to Water Quality**

When added to natural water, NaOCl reacts with a wide variety of substances, including ammonia and naturally occurring humic materials. The ammonia reacts with HOCl to form chloramines, which have significant disinfecting power and may serve to impair the dechlorinating population during bioremediation. If there is significant natural organic matter present, halogenated disinfection by-products (such as chloroform) may be formed (Sawyer et al, 1994).

### **5.2.4 Implementation Issues**

NaOCl is widely used in well rehabilitation applications and should be widely accepted by regulators. Although storage and dosage are simple, NaOCl is a corrosive substance at high concentrations and should be handled with care.

## **5.3 GLYCOLIC ACID**

### **5.3.1 Mode of Action**

Glycolic acid is a chelating agent that binds with divalent cations in the biofilm matrix, reducing its mechanical strength and facilitating biofilm detachment at lower shear stresses.

In groundwater environments, the use of organic acids, such as glycolic acid, is particularly attractive given the relative biodegradability of these compounds and the accompanying beneficial decrease in pH that will promote the removal of carbonate minerals contained within the biofilm. For well rehabilitation, chelating or dispersing agents, such as glycolic acid, are used to detach the biofilm from the well screen and filter pack, and the detached biomass is then pumped out of the well bore. Used in a preventative strategy along with nutrient amendment, a dispersing or chelating agent could be co-amended with the nutrient, inhibiting biomass attachment and preventing initial biofilm formation.

### **5.3.2 Where Used**

Glycolic acid is widely used in the groundwater industry for water well rehabilitation, and glycolic acid has also been used preventatively for biofouling control. Commercial glycolic acid products include LBA (CETCO). LBA is a (formerly) National Sanitation Foundation (NSF) Drinking Water Additive (NSF 60)-registered product.

### **5.3.3 Impacts to Water Quality**

Limited data on the impacts of glycolic acid on groundwater quality and the microbial community structure are available. A reduction in pH will occur, which could raise the solubility of some mineral phases. Field data are required to better understand the potential impacts of this biofouling control on groundwater quality when used in a preventative capacity.

### **5.3.4 Implementation Issues**

The use of glycolic acid raises no known implementation issues other than conventional federal or state user identification code (UIC) permitting. As a result of its existing commercial availability, it is generally widely accepted by the consulting and regulatory communities. Existing commercial products are NSF-60 registered, indicating that their presence in drinking water is considered acceptable.

## **5.4 AQUA GARD™ PROCESS**

### **5.4.1 Mode of Action**

Liquid and gaseous carbon dioxide are injected into the well periodically through permanent placement of injection equipment, as shown in Figure 4. The carbon dioxide undergoes a phase change resulting in a mechanical scouring action, which removes inorganic scale and biofouling. Debris is pumped off using an airlift pump.

### **5.4.2 Where Used**

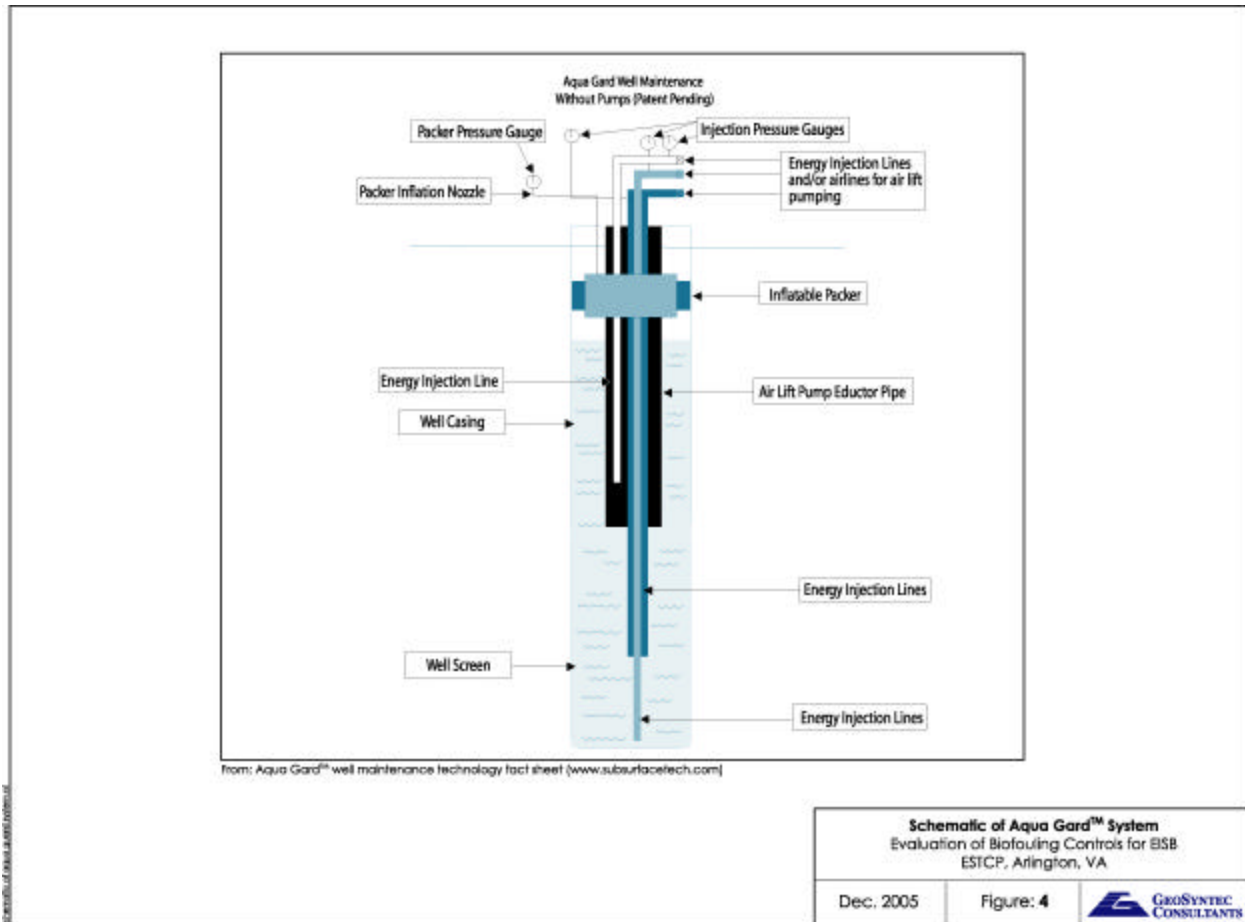
The Aqua Gard™ process has been developed as a biofouling control process for water wells. A related process, Aqua Freed™, is used for well rehabilitation.

### **5.4.3 Impacts to Water Quality**

Addition of carbon dioxide can act to lower the pH of the groundwater through the formation of carbonic acid.

### **5.4.4 Implementability Issues**

This technology requires that the well be specially designed to permit the separate addition of carbon dioxide and removal of debris. Currently, a licensed contractor is required to perform the treatments. The cost of the technology is reasonable, providing a contractor is located near the EISB site. This technology is commercially available and should pose no regulatory hurdles as



**Figure 4. Schematic of Aqua Gard™ System.**

only carbon dioxide is added to the aquifer. An automated version of this technology is in development.

## 5.5 THERMAL PASTEURIZATION

### 5.5.1 Mode of Action

Heat is added to kill the bacteria in the biofilm. Water heated to 54°C and recirculated within the well over several days has been shown to be effective in dispersing clays and treating biofouling in the short term (Smith, 1995). For biofouling control applications, heat could be applied periodically to minimize biofouling in the well and filter pack by periodically adding hot water. Heat addition can also improve the effectiveness of chemical well rehabilitation treatments, as in the case of the BCHT process (U.S. Patent 4,765,410, ARCC Inc., Dayton Beach, Florida).

### 5.5.2 Where Used

Thermal pasteurization has been used for well rehabilitation. Pasteurization has been used extensively in the food and beverage industry to significantly reduce the number of bacteria in food and beverages, such as milk.



### **5.5.3 Impacts to Water Quality**

Heat treatment has the advantage that no chemicals are added to the aquifer. However, heating may decrease the solubility of carbonates in the aquifer.

### **5.5.4 Implementability Issues**

Aquifer materials subject to regular heating can store heat, which can cause grout to dry out and crack. Excessive heat is not favourable for most plastic components used in wells, but temperatures applied at less than 60°C in the well are within the tolerance of polyvinyl chloride (PVC) casing (Smith, 1995). If misapplied, heating can encourage growth at the edge of the thermal impact zone and possibly increase biofouling within the aquifer (Smith, 1995).

## 6.0 CONCLUSIONS

Biofouling is a common occurrence in injection wells used for EISB applications because the bioremediation process adds nutrients directly to the injection well(s), creating conditions favorable for microbial growth and biofilm formation. Currently, there are few proven preventative biofouling controls for EISB systems. Several conventional well rehabilitation methods exist, but they are costly, involving significant system downtime and often the use of service rigs and biocidal chemicals, which require handling precautions. Furthermore, biofouling usually returns within a relatively short time frame (e.g., months), requiring subsequent rehabilitation events. The use of biofouling controls represents a way to minimize or eliminate biofouling and well rehabilitation, increasing the cost-effectiveness and performance of EISB systems.

There are several general categories of biofouling controls, including oxidizing biocides, nonoxidizing biocides, dispersing and chelating agents, physical and thermal approaches. The ideal biofouling control approach provides the requisite level of biofouling control within the nutrient delivery wells and filter pack, minimizes adverse environmental or toxic and geochemical impacts, requires a low dose, is cost-effective, is easily automated, and can achieve regulatory acceptance.

As a result of this review, the following measures were identified as promising biofouling controls for groundwater applications: chlorine dioxide, NaOCl, glycolic acid, the addition of carbon dioxide (Aqua Gard™), and thermal pasteurization. A field demonstration/validation program will be implemented in 2006 to evaluate these promising biofouling controls head-to-head, along with two experimental controls without biofouling controls (one with and one without electron donor). Time and budget permitting, other biofouling controls such as polymaleic acid, citric acid, and other chlorine dioxide processes, may also be evaluated.

*This page left blank intentionally.*

## 7.0 REFERENCES

1. Agency for Toxic Substances and Disease Registry (ATSDR). 2002. Sodium Hypochlorite Fact Sheet. CAS #7681-52-9. April 2002.
2. Alford, G. and R. Cullimore. *The Application of Heat and Chemicals in the Control of Biofouling Events in Wells*. Lewis Publishers, Boca Raton, Florida. 1999.
3. ASTM D 3623-78a, *Standard Test Method for Testing Antifouling Panels in Shallow Submergence*. ASTM International. 1998.
4. Australian Drilling Industry Training Committee Limited (ADITC). *Drilling. The Manual of Methods, Applications, and Management*. J.S. McMillan Printing Group, Australia. 2002.
5. Baveye, P., P. Vandevivere, B.L. Hoyle, P.C. DeLeo, and D.S. de Lozada. Environmental impact and mechanisms of the biological clogging of saturated soils and aquifer materials. *Critical Reviews in Environmental Science and Technology*. 28(2):123-191. 1998.
6. Camper, A., M. Burr, B. Ellis, P. Butterfield, and C. Abernathy. Development and Structure of Drinking Water Biofilms and Techniques for Their Study. *J. Appl. Microbiol. Symposium Suppl.* 85:1S-12S. 1999.
7. Characklis, W.G., and K.C. Marshall. *Biofilms*. John Wiley and Sons, Inc, New York. 1990.
8. Cloete, T.E., V.S. Brozel, and A. von Holy. Practical aspects of biofouling control in industrial water systems. *International Biodeterioration & Biodegradation*. 29:299-341. 1992.
9. Cox, E.E., C. Elder, E. Hood, E. Edwards, J. Klaiber, B. Smith, and J. Berry. In Situ Bioremediation of Chlorinated and Non-Chlorinated VOCs in Landfill Groundwater Using a Sulfate Biobarrier. Presented at the *Third International Symposium on Remediation of Chlorinated and Recalcitrant Compounds*. Monterey, California. May 2002a.
10. Cox, E.E., R. Borch, T. McAlary, M. McMaster, S. Neville, and L. Bonsack. In Situ Bioremediation of Perchlorate: Comparison of Results from Multiple Field Demonstrations. Presented at the *Third International Symposium on Remediation of Chlorinated and Recalcitrant Compounds*. Monterey, California. May 2002b.
11. Cox, E.E., R. Borch, and S. Neville. A Survey of In Situ Bioremediation Applications for the Reduction of Perchlorate in Soil and Groundwater. In: *Proceedings of the Groundwater Resources Association Conference on Perchlorate in Groundwater: Occurrence, Analysis and Treatment*. Sacramento, California, July 31, 2003.

12. Cullimore, R. *Microbiology of Well Biofouling*, Lewis Publishers, CRC Press, Boca Raton, Florida. 1999.
13. Devlin, J.F., and D. Muller. Field and Laboratory Studies of Carbon Tetrachloride Transformation in a Sandy Aquifer under Sulfate Reducing Conditions. *Environmental Science & Technology*. 33(7):1021-1027. 1999.
14. Dybas, M.J., M. Barcelona, S. Bezborodnikov, S. Davies, L. Forney, H. Heuer, O. Kawka, T. Mayotte, L. Sepulveda-Torres, K. Smalla, M. Sneathen, J. Tiedje, T. Voice, D.C. Wiggert, M.E. Witt, and C.S. Criddle. Pilot-Scale Evaluation of Bioaugmentation for In Situ Remediation of A Carbon Tetrachloride-Contaminated Aquifer. *Environmental Science & Technology*. 32(22):3598-3611. 1998.
15. Elimelech. M., J. Gregory, X. Jia, and R. Williams. *Particle Deposition and Aggregation: Measurement, Modeling and Simulation*. Butterworth-Heinemann, Newton, Massachusetts. 1995.
16. Ellis, D.E., E.J. Lutz, J.M. Odom, R.J. Buchanan, C.L. Bartlett, M.D. Lee, M.R. Harkness, and K.A. DeWeerd. Bioaugmentation for accelerated in situ anaerobic bioremediation. *Environmental Science & Technology*. 34(11):2254-2260. 2000.
17. Fry, N.K., J.K. Fredrickson, S. Fishbain, M. Wagner, and D.A. Stahl. Population structure of microbial communities associated with two deep, anaerobic, alkaline aquifers. *Applied and Environmental Microbiology*. 63(4): 1498-1504. 1997.
18. Gates, D. *The Chlorine Dioxide Handbook*. American Water Works Association. U.S.A. 1998.
19. Geosyntec Consultants, Inc. *Pilot Test of In Situ Bioremediation of Perchlorate and Trichloroethene in Groundwater* Report. Inactive Rancho Cordova Test Site, Western Non-Aerospace, Non-Industrial Area. Prepared for Aerojet General Corporation and The Boeing Company. August 12, 2004.
20. Geosyntec Consultants, Inc. 2003. *Pilot Test For In Situ Bioremediation of Perchlorate-Impacted Groundwater*. Final Report. Former Pepcon Facility, Henderson, Nevada. American Pacific Corporation. May, 2003.
21. Globa, R, and H. Rohde. Application of Impressed Current Systems to Mitigate Water Well Biofouling. 56th Canadian Geotechnical Conference, 4th Joint IAH-CNC/CGS Conference, 2003 NAGS Conference. 2003.
22. Griffiths, J.R. *Nontoxic Antifouling Systems*. U.S. Patent No, 5,449,553, 1995.
23. Griffiths, J.R. *Nontoxic Antifouling Systems*. U.S. Patent No, 5,593,732, 1997.
24. Helweg, O.J., V.H. Scott, and W.C. Scalmanini. *Improving Well and Pump Efficiency*. American Water Works Association. Denver, Colorado. 1983.

25. Lendvay, J.M., F.E. Loffler, M. Dollhopf, M.R. Aiello, G. Daniels, B.Z. Fathepure, M. Gebhard, R. Heine, R. Helton, J. Shi, R. Krajmalnik-Brown, C.L. Major, M.J. Barcelona, E. Petrovskis, R. Hickey, J.M. Tiedje, and P. Adriaens. Bioreactive Barriers: A Comparison of Bioaugmentation and Biostimulation for Chlorinated Solvent Remediation. *Environmental Science & Technology*. 37(7):1422-1431. 2003.
26. Liikanen, R., J. Yli-Kuivila, and R. Laukkanen. Efficiency of various chemical cleanings for nanofiltration membrane fouled by conventionally treated surface water. *Journal of Membrane Sciences*. 195:265-276. 2002.
27. Major, D.W., M.L. McMaster, E.E. Cox, E.A. Edwards, S.M. Dworatzek, E.R. Hendrickson, M.G. Starr, J.A. Payne, and L.W. Buonamici. Field Demonstration of Successful Bioaugmentation to Achieve Dechlorination of Tetrachloroethene to Ethene. *Environmental Science & Technology*. 36(23):5106-5116. 2002.
28. Mansuy, N. *Water Well Rehabilitation, A Practical Guide to Understanding Well Problems and Solutions*, CRC Press Lewis Publishers, Boca Raton, FL. 1999.
29. Mansuy, N. *Aqua Gard™ Permanent In-Well Maintenance Devices Significantly Improve Well Performance and Reduce Well Maintenance Costs*. 2003.
30. Mayer, C., R. Moritz, C. Kirschner, W. Borchard, R. Maibaum, J. Wingender and H. Flemming. The Role of Intermolecular Interactions: Studies on Model Systems for Bacterial Biofilms. *Int. J. Bio. Macromol.* 26: 3-16. 1999.
31. McCarty, P.L., M.N. Goltz, G.D. Hopkins, M.E. Dolan, J.P. Allan, B.T. Kawakami, and T.J. Carrothers. Full-Scale Evaluation Of In Situ Cometabolic Degradation of Trichloroethylene in Groundwater through Toluene Injection. *Environmental Science & Technology*. 32(1):88- 100. 1998.
32. Millar, K., S. Lesage, S. Brown, C.S. Mowder, T. Llewellyn, S. Forman, D. Peters, G. DeLong, D.J. Green, and H. MacIntosh. "Biocide Application Prevents Biofouling of a Chemical Injection/Recirculation Well", In the proceedings of the *Sixth International Symposium on In Situ and On-Site Bioremediation*, San Diego, California; Battelle Press, pp. 333-348. 2001.
33. Montgomery, J.M. *Water Treatment Principles and Design*, Wiley Interscience. 1985.
34. Neu, T.R., H.C. van der Mei, and H.J. Busscher. Biofilm associated with health: in *Biofilms - Science and Technology*, L.F. Melo, T.R. Bott, M. Fletcher, and B. Capdeville (eds), Dordrecht, Kluwer Academic Publishers, 21-34. 1992.
35. Noss, C.I., W.H. Dennis, and V.P. Olivieri. Reactivity of Chlorine Dioxide with Nucleic Acids and Proteins. *Water Chlorination: Environmental Impact and Health Effects*. R.L. Jolley et al, (Eds), Lewis Publishers. 1983.

36. Parkar, S.G., S.H. Flint, and J.D. Brooks. Evaluation of the effect of cleaning regimes on biofilms of thermophilic bacilli on stainless steel. *Journal of Applied Microbiology*. 96:110-116. 2004.
37. Sawyer, C. N., P. L. McCarty, and G. F. Parkin. *Chemistry for Environmental Engineering*, Fourth Edition. McGraw-Hill, Inc., New York. 1994.
38. Smith, S.A. *Monitoring and Remediation of Remediation Wells. Problem Prevention, Maintenance, and Rehabilitation*. CRC Press, Lewis Publishers, Boca Raton, Florida. 1995.
39. Taylor, S.W., and P.R. Jaffe. Enhanced In-Situ Biodegradation and Aquifer Permeability Reduction. *Journal of Environmental Engineering*. 117(1): 25-46. 1991.
40. Urfer, D., P.M. Huck, S.D.J. Booth, and B.M. Coffey. Biological Filtration for BOM and Particle Removal: A Critical Review. *Journal of the American Water Works Association*. 89(12): 83-98. 1997.
41. U.S. Army Corps of Engineers (USACE). *Engineering and Design Operation and Maintenance of Extraction and Injection Wells at HTRW Sites*. Pamphlet No. 1110-1-27. 27 January 2000.
42. U.S. Environmental Protection Agency. 1999. *Alternative Disinfectants and Oxidants*. Guidance Manual. EPA 815-R-99-014.
43. U.S. Environmental Protection Agency. *Sodium and Calcium Hypochlorite Salts: Reregistration Eligibility Decision (RED) Fact Sheet*. EPA 738-F-91-108, September 1991.
44. Werdehoff, K.S., and P.C. Singer. Chlorine Dioxide Effects on THMFP, TOXFP, and the Formation of Inorganic By-Products. *J. AWWA*. 79(9):107. 1987.
45. Zinn et al. Environmentally acceptable control of microbial biofilms, in *Biofilms: Recent Advances in Their Study and Control*. L.V. Evans, editor. Amsterdam, Harwood Academic Publishers, pp. 19-34. 2000.