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Citation: Li, Kwan, Matthew Whitfield, and Krystyn J. Van Vliet. "Beating the bugs: roles of microbial biofilms in corrosion." *Corrosion Reviews* 321, 3-6 (2013); © 2013, by Walter de Gruyter Berlin Boston. All rights reserved.

As Published: <https://dx.doi.org/10.1515/CORRREV-2013-0019>

Publisher: Walter de Gruyter GmbH

Persistent URL: <https://hdl.handle.net/1721.1/125679>

Version: Author's final manuscript: final author's manuscript post peer review, without publisher's formatting or copy editing

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Beating the bugs: Roles of microbial biofilms in corrosion

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Abstract

Microbiologically influenced corrosion is a complex type of environmentally assisted corrosion. Though poorly understood and challenging to ameliorate, it is increasingly appreciated that MIC accelerates failure of metal alloys, including steel pipeline. Historically, this type of material degradation process has been treated from either an electrochemical materials perspective or a microbiological perspective. Here, we review the current understanding of MIC mechanisms for steel – particularly those in sour environments relevant to fossil fuel recovery and processing – and outline the role of the bacterial biofilm in both corrosion processes and mitigation responses.

Keywords: biofilm; sulfate-reducing bacteria (SRB); microbiologically influenced corrosion (MIC)

1 Introduction

Microbiologically influenced corrosion (MIC) can accelerate mechanical failure of metals in a wide range of environments ranging from oil and water pipelines and machinery to biomedical devices. Though this mode of environmentally assisted corrosion has no single definition, MIC invariably includes corrosion in the presence of microbial species including bacteria and the associated biofilms produced by those bacteria. MIC has been noted to occur for a variety of metals and alloys such as iron and steel, copper, and titanium [Rao, 2012], as well as on nonmetallic materials such as concrete [George, 2012]. For instance, MIC and associated biofouling are estimated to account for 20% of direct fuel pipeline integrity and reliability costs exceeding \$2 billion USD per year [Flemming, 1996, Koch et al., 2002]. Recently, this corrosion mechanism has been implicated in several rapid, high-profile failures of pipeline including a light grade crude oil pipeline failure [Al-Jaroudi et al., 2011], a seawater pipeline failure within a few years of installation [Abraham et al., 2009],

leaks in stainless steel cooling water system [Yu et al.,], and leaks in the Trans-Alaskan Pipeline [Egan, 2011].

Despite decades of research, the exact mechanisms by which and environments in which MIC enhances corrosion are poorly understood. Indeed, even within the same oilfield or pipeline, some locations may exhibit MIC while others do not [Yu et al., , Voordouw et al., 2009, Maruthamuthu et al., 2011]. Conventionally, this phenomenon has been studied either from a biological perspective – considering the complex nature of bacteria consortia and biofilm formation – or from a materials corrosion perspective – considering the evidence and mitigation of metal pitting under controlled conditions. Therefore, there is no consensus on how to mitigate corrosive losses due to MIC. Here, we review the most current understanding of MIC mechanisms on steel (i.e., iron-carbon or ferrous alloys), MIC mitigation strategies, and the role of the bacterial biofilm in both corrosion and mitigation response.

2 Characteristics of MIC

MIC can occur in both aerobic or anaerobic conditions, and is defined by no single characteristic trait other than corrosion in the presence of microbial species (bacteria) and their associated biofilms. Thus, MIC is considered biotic corrosion, and the terms "microbes" and "bacteria" are used interchangeably. As bacteria are ubiquitous in almost all environments (air, water, and organic fluids), the mere presence of bacteria found at sites of corrosion does not necessarily mean that the bacteria were a significant contributing factor in corrosion initiation or acceleration. MIC can occur in conjunction with other types of abiotic corrosion, further confuscating attempts to identify the root cause of corrosion failure. Often, MIC is associated with surface pitting, which leads to more rapid corrosive failure than uniform corrosion [Bryant et al., 1991, Chamritski et al., 2004, Jan-Roblero et al., 2004, Miranda et al., 2006, Yuan and Pehkonen, 2009] (Figure 1). Thus, even nonferrous metals that exhibit a naturally passivating layer at the surface (e.g., copper oxides and titanium oxides) exhibit MIC when metabolic byproducts of the microbes serve to chemically reduce and thus disrupt the passivating film under anaerobic conditions; this results in pitting corrosion at the metal-biofilm interface [Rao, 2012]. As many surface characteristics including roughness, charge, and hydrophilicity can modulate bacterial adhesion, it is not yet clear whether bacteria initiate the pits or adhere preferentially to pre-existing pits. Although several studies have found evidence that microbes adhere preferentially to anodic metal regions such as welded joints, scratched edges, grain boundaries [Dexter and Eashwar, 1999, Sreekumari et al., 2001], or to previously corroded locations [Little et al., 1999], a more recent study was not able to confirm these site-specific claims [Sherar et al., 2011]. When many species of bacteria adhere and proliferate on a surface, they secrete a polysaccharide-rich matrix; a biofilm includes that extracellular polysaccharide matrix and the bacterial cells within. Attempts to spatially correlate localized corrosion with the

overlying biofilm, e.g., using electrochemical mapping techniques, have failed due to the conductive nature of the biofilm itself [Dong et al., 2011]. Further, although pitting is a known signature of MIC, others have found no unique characteristics that distinguish the morphology of MIC pits from abiotic pits [Thomas and Chung, 1999].

[Figure 1 positioned here.]

At sites of suspected MIC, a so-called consortium of several bacterial species is always found – rather than a single species – though the number and types of bacteria in that consortium varies widely from location to location. Because the culture conditions of the majority of bacterial species are not well-established, determining the phylogenetic makeup is difficult. However, the application of molecular techniques now allow much more detailed analysis of extracts, quantifying the types and relative numbers of species [Amann et al., 1995]. Several studies have performed detailed phylogenetic characterization of fluids from gas and oil pipelines [Jan-Roblero et al., 2004, Rajasekar et al., 2009, Maruthamuthu et al., 2011] and water outlet pipes at oil production facilities [Larsen et al., 2010]. The categories of bacteria typically directly linked to corrosion are sulfate-reducing bacteria (SRB), iron oxidizing bacteria, sulfur oxidizing bacteria, nitrate reducing bacteria (NRB), and methanogens. These bacteria may reduce the metal directly, produce corrosive metabolic byproducts, and/or produce biofilms that indirectly alter the local environment to promote corrosion [Rao, 2012].

SRB are considered one of the main culprits of biotic corrosion in anaerobic conditions. SRB are a grouping of bacteria that includes at least 220 species which produce H_2S , and use sulfates as the terminal electron acceptor [Barton and Fauque, 2009]. Most SRB are considered obligate anaerobes, meaning that the cells cannot metabolize and/or replicate in the presence of oxygen, although many species can temporarily tolerate low levels of oxygen [Johnson et al., 1997]. Furthermore, anaerobic conditions capable of supporting SRB growth can be created in overall aerobic environments, due to the microniches created within the bacterial biofilm/corrosion product layer. Although SRB are the most studied and well understood of the anaerobic corrosion inducing bacteria, we note that MIC can occur in anaerobic conditions in the absence of SRB [Rajasekar et al., 2009]. However, given the wide acknowledgment of SRB presence in industrial MIC contexts, and the substantial study of this bacteria grouping at the genetic and structural levels [Wall et al., 2008], we will focus primarily on the role of this microbe type within the consortium and biofilms relevant to MIC of ferrous alloys.

3 Characterisitics of MIC-Associated Biofilms

MIC is attributed not to planktonic bacteria, but instead to adherent bacteria in the form of biofilms on surfaces. Many bacteria types adhere to and form polysaccharide-rich biofilms on material surfaces, with interactions governed strongly by the material surface properties,

fluid properties, and bacteria adhesion mechanisms [Lichter et al., 2008, Lichter et al., 2009]. Biofilm formation follows a series of critical steps: an initial rapid and reversible bacterial attachment, a more stable, longer term attachment, bacterial replication and matrix secretion, biofilm maturation, and finally bacterial dispersal [Lichter et al., 2009, Monds and O’Toole, 2009]. The main structural components of the biofilms are produced by the bacteria themselves through the secretion of extracellular polymeric substances (EPS). EPS is composed chiefly of polysaccharides, proteins, nucleic acids, and lipids [Vu et al., 2009]. Biofilms can be anywhere from μm to mm in thickness. These structures can develop and mature over the course of hours, days, or months, depending on the bacterial species and environment (Figure 2). Repeated biofilm formation has the potential to occur due to pigging, an intentional mechanical delamination of corrosion products including iron sulfides within internal pipeline surfaces, or due to general biofilm rupture. In the context of fuel transport within pipelines, the sporadic flow rates and fluid composition can also disrupt and promote biofilm formation, and likewise the presence of the biofilm can affect flow trajectories. Essentially, through these biofilms the bacteria create a unique and sustaining microenvironment that can differ significantly – in terms of composition and distribution of solids, fluids, and gases – from the overall macroenvironment.

[Figure 2 positioned here.]

The formation of a biofilm itself does not necessitate an increased corrosion rate, and can actually lead to corrosion resistance in some circumstances. Certain bacteria within biofilms can remove corrosion-promoting agents (such as O_2 [Dubiel et al., 2002]) as part of the bacteria metabolism, can secrete antimicrobial agents that inhibit growth of corrosion causing bacteria, and can generate physical barriers that protect surfaces from corrosion [Zuo, 2007, Avidan et al., 2010]. The particular species makeup and environmental conditions greatly influence the corrosive potential of a biofilm. This variation has obfuscated identification of a general, causal link between bacteria presence and corrosion. Such complexity prohibits the community from drawing broad conclusions on corrosion potential, mechanisms, and rates beyond the specific conditions in which observations were made. This difficulty has prompted the development of models to better predict when MIC will be a problem [Maxwell, 2006, Skovhus et al., 2012], but the applicability of such models is still limited currently to specific species and environmental conditions.

While single-species studies are naturally better controlled and simpler to interpret, recognition of the significance of the interplay among the different bacterial species within a biofilm has prompted multiple-species studies. For instance, Okabe et al. studied the spatial distribution of SRB in an aerobic wastewater biofilm and found that although SRB were located throughout the biofilm, sulfate reduction was focused in a narrow anoxic band [Okabe et al., 1999]. Andersson et al. systematically studied initial adherence and biofilm formation with different combinations of 13 bacterial species often found in wastewater systems, and found complex protaganistic and antagonistic interactions that depended on

species composition [Andersson et al., 2008]. Interestingly, Lee et al. found that a dual-species biofilm with an SRB and iron-reducing bacteria resulted in reduced corrosion, as compared to a biofilm comprising only SRB [Lee et al., 2006]. Videla et al. also found differences in corrosion with iron- and sulfate-reducing bacteria in mixed culture in different environments [Videla et al., 2008]. Overall, however, these studies illustrate the complexity of the process, rather than providing concrete conclusions about MIC mechanisms.

Environmental factors such as fluid shear flow found within pipelines add another layer of complexity to biofilm development, and thus to MIC in industrial settings. It is known that shearing forces can greatly influence bacterial adhesion and biofilm development in dramatic ways, regardless of the context [Rochex et al., 2008, Whitfield et al., 2010]. For example, in other bacterial species (aerobic species such as *E. coli*), it has been shown that adhesion is strengthened in the presence of moderate flow rates. At lower rates, the so-called catch bond mechanism is not activated, and at higher rates the binding is again disrupted; only at intermediate rates are the adhesive structures at the bacterial surface configured to bind to the surface with highest affinity and longest lifetime [Thomas et al., 2002, Whitfield et al., 2010]. For SRB, it is unknown whether similar adhesive structures and mechanisms exist. However, moderate flow rates (0.1-0.5 m/s) have been shown to change SRB biofilm morphology, and to increase biofilm rigidity and yield shear stress [Stoodley et al., 2001, Dunsmore et al., 2002]. At sufficiently high flow rates (3.5 m/s), SRB adhesion and biofilm growth are inhibited [Wen et al., 2007]. Qualitatively, these results are similar to more widely studied aerobic biofilms, making it possible that mechanistic insights can be shared. In a study not directly related to corrosion research, Rochex et al. found that increased shear stresses lowered the species diversity within biofilms [Rochex et al., 2008]. A more robust understanding of how flow conditions affect MIC-associated biofilm structure, diversity, and corrosive potential would allow for improved a risk assessment and pipeline flow rate and flow pattern selection.

4 Mitigation Techniques

A number of techniques have been investigated to mitigate the effects of MIC. These include using corrosion resistant metals, protective coatings, pigging, anodic and cathodic protection, biocides, introduction of competing bacteria, and nutrient addition or removal. Some techniques such as pigging (i.e., mechanical delamination of corrosion products) and biocide addition are widely used in field conditions that include sour (anaerobic, sulfur-containing) environments such as buried pipeline. Other approaches such as sol-gel coating barriers [Akid et al., 2008] or precoating with purified EPS [Stadler et al., 2010] are still in experimental stages. One of the more recently field tested techniques - and one that relies on a robust understanding of the biofilm - is to alter the corrosive ability of biofilms by manipulating the available nutrients via addition of nitrates or removal of sulfates.

In field conditions, nitrate addition [Voordouw et al., 2009] and sulfate removal [McEl-

hiney et al.,] have been shown to affect SRB activity and be somewhat effective in reducing or altering corrosion. Nitrate addition can allow other bacteria such as nitrate reducing bacteria to outcompete the more corrosive SRB (Figure 3)[Thorstenson et al., 2002, Hubert and Voordouw, 2007]. Additionally, there is evidence that nitrate can directly inhibit the SRB metabolism and proliferation [Haveman et al., 2004, He et al., 2010]. However nitrate addition is not a definitive solution to MIC. Although this approach typically reduces sulfide production, the effect is only temporary and SRB are not killed. Thus, sulfide production rapidly returns if active nitrate treatment is terminated [Mohanakrishnan et al., 2011]. Another study showed that nitrate addition could reduce uniform surface corrosion but not pitting corrosion, the latter of which is often the cited cause of pipeline failures [Schwermer et al., 2008]. Further, the same nutrient manipulation protocols work in some pipelines but not others, necessitating trial and error at each location to achieve the best results [Voordouw et al., 2009]. One study even found that nitrates can increase corrosion of steel [Marques et al., 2012]. Finally, even when corrosion inhibition is achieved, for applications such as fuel recovery and processing, the addition of such chemicals then requires subsequent removal processes for the fluid of interest; this is not a treatment localized to the metal surface, and thus the high-volume addition and removal of chemical agents affects product cost and thus the decisions to employ such anti-MIC strategies. Thus, while nutrient alteration has produced intriguing results on MIC mitigation, it has yet to produce a definitive solution.

[Figure 3 positioned here.]

5 Mechanisms of MIC

Owing to the complexity of the microbe-metal interaction, the underlying mechanism of MIC remains an open question and subject to much debate. Many possible mechanisms for MIC have been proposed, and it is possible that no predominant mechanism exists. In fact, this seems likely given the incredible diversity of species and conditions in which MIC is observed. It is known that chemical, mechanical and structural factors all play a role, both biotic and abiotic in nature, and these may all work in concert to cause and sustain MIC [Beech and Gaylarde, 1999]. Thus, it must be emphasized that evidence for one hypothesis is not necessarily contradictory to another. The given environmental conditions and bacterial species present may dictate the predominant mechanism.

Among the first proposed mechanisms for MIC, the cathodic depolarization theory - also known as the classical theory - was put forth in 1934 by Kuhr et al. in order to explain the unexpectedly high rate of corrosion failures encountered on buried cast iron pipelines in the Dutch countryside [von Wolzogen Kuehr and Van der Vlugt, 1964]. SRB were identified as the culprit due to the ferrous sulfide corrosion products found in conjunction with these failures. In anaerobic conditions where SRB thrive, hydrogen ions typically serve as the terminal electron acceptor at the cathode in a corrosion reaction. This reduced hydrogen

would then adsorb onto the metal surface, polarizing it. According to the classical theory, the role of the SRB was to consume this cathodic hydrogen by means of the enzyme hydrogenase, catalyzing the recombination of the adsorbed atomic hydrogen into hydrogen gas, thus depolarizing the cathode.

Though the classical theory was grounded in electrochemistry and found general acceptance for nearly half a century, it was ultimately overturned when new techniques showed that hydrogenase can act only on molecular hydrogen, not atomic hydrogen [Stott, 1993]. In support of this finding, Cord-Ruwisch et al. showed that nitrate-reducing bacteria with an enhanced ability to consume hydrogen failed to cause noticeable corrosion [Cord-Ruwisch, 1996]. Despite these findings, however, research continues into the role of hydrogenase in MIC, often with contradictory results. Studies by Bryant et al. [Bryant et al., 1991] and Da Silva et al. [Da Silva et al., 2002] showed a positive correlation between the presence of hydrogenase and MIC, while King et al. [King et al., 1973b] found no such link.

Following the fall of the classical theory, myriad “alternative theories” have been proposed, often attempting to explain MIC while minimizing the role of the bacteria themselves [Javaherdashti, 2011]. Such theories include anodic depolarization [Crolet, 1992], the presence of a volatile phosphorous compound [Iverson and Olson, 1983], metal-binding exopolymers [Beech and Cheung, 1995], sulfide-induced stress corrosion cracking and hydrogen-induced blistering [Edyvean et al., 1998]. Here, however, we restrict our focus to the biofilm and its role in MIC. Figure 4 illustrates the mechanisms by which a biofilm can contribute to corrosion initiation and accelerated corrosion rates.

5.1 The biofilm as a chemical barrier

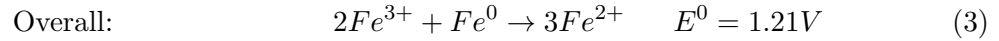
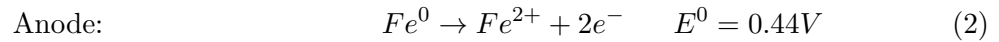
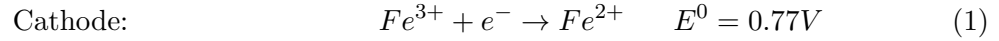
[Figure 4 positioned here.]

The existence of a biofilm can impede the diffusion of chemical species – both into and out of the biofilm – producing localized chemical environments that are significantly different from the bulk [Beech and Sunner, 2004]. These localized environments can, in turn, greatly affect the underlying corrosion in a number of ways (Figure 4). In aerobic conditions, the oxygen concentration may differ greatly across the biofilm due both to O_2 diffusion limitations as well as O_2 consumption by aerobic bacteria. Thus, even in an aerated environment, anaerobic regions can develop beneath a biofilm allowing for the proliferation of SRB and other corrosion-enhancing anaerobes (Figure 4A). By inhibiting the diffusion of corrosive chemicals away from the metal surface, the biofilm can effectively act as a concentration multiplier. The H_2S generated by SRB in anaerobic regions is potentially much more potent when encapsulated beneath a biofilm than otherwise (Figure 4B). King et al. demonstrated a significant difference in the rate of corrosion between an SRB colony and a sterile medium to which an equivalent quantity of sulfide was periodically added [King et al., 1973b]. In the former case, the biogenic sulfides were produced beneath a biofilm and near the metal surface, while in the latter case the abiotic sulfide was added directly to the bulk solution.

In addition to concentrating the H_2S , the biofilm can also concentrate the corroded ferrous ions, a key factor in MIC (Figure 4C). Past studies [King et al., 1973b, Lee and Characklis, 1993] have shown that the presence of ferrous ions is a major factor for MIC to occur on iron or steel in an aqueous environment. In [Lee and Characklis, 1993], Lee et al. grew a pure culture of SRB in the absence of ferrous ions and little to no corrosion was observed despite a high level of microbial activity. Table 1 demonstrates the positive correlation between ferrous ion concentration and corrosion rates. This strong correlation implies that ferrous ions play an integral part in the corrosion mechanism of MIC, at least in the presence of SRB. Two possible roles may be suggested: that the ferrous/ferric ion ($\text{Fe}^{2+}/\text{Fe}^{3+}$) redox couple plays a significant role, and/or the ferrous corrosion products do. It should be mentioned that ferrous ions are not known to affect the metabolism of SRB in any manner, though other bacteria are known to use them to cause corrosion on metals other than steel, such as *Acidithiobacillus ferrooxidans* in the industrial process of bioleaching [Rohwerder et al., 2003].

[Table 1 positioned here.]

In a metal/solution interface between iron and aqueous ferrous and ferric ion, a galvanic cell is established and the following two half-cell reactions take place:



where the standard electrode potentials are referenced to the standard hydrogen electrode. When bound to functional groups in the biofilm, these potentials may become even more conducive toward corrosion, as previously discussed. Nevertheless, as an oxidant, ferric ions ($E^0 = 0.77V$) are more reducing than hydrogen ($E^0 = 0V$) but less than oxygen ($E^0 = 1.23V$), meaning that at the same ion concentrations, a cathodic reaction involving oxygen would be preferred. However, the strength of ferric ions as an oxidant in the corrosion of iron or steel is a positive feedback loop, as illustrated in Figure 4C. Unlike in the oxygen reduction reaction wherein the reactant oxygen molecules are continually consumed - potentially leading to diffusion limiting kinetics - ferrous ions are continually regenerated as this corrosion reaction proceeds. Moreover, each new ferrous ion is generated at the interface, preventing the development of diffusion limited kinetics. These ferrous ions can then be oxidized by oxygen to restore the ferric ion concentration. For $\text{pH} > 4.5$, the rate of this reaction is known to be very fast and thus does not limit the rate of corrosion [Singer et al., 1970].

The presence of the biofilm aids this mechanism in two ways: first, it further concentrates the ferric ions near the metal surface, and second it spatially separates the two

oxidation reactions [Beech and Sunner, 2004]. Figure 4D illustrates the second point: electrons lost by ferrous ions in the anaerobic zone can travel through the conductive biofilm to be accepted by oxygen in the aerobic zone. By removing the need for either species to diffuse through the biofilm, the rate of the corrosion reaction can thus be greatly increased. This suggests that the presence of the biofilm, rather than the presence of the bacteria that made and stabilized that biofilm, directly increases corrosion rate. Separately, we note that species of iron-oxidizing bacteria, which as part of their metabolism oxidizes Fe^{2+} to Fe^{3+} , have been identified at MIC events, and these have also been implicated as a potential cause for MIC [Rajasekar et al., 2010].

5.2 Physical structure of the biofilm

The physical structure and heterogeneous nature of the biofilm also contribute to the corrosion process. Biofilms are often patchy and of varying thickness at the microscale, creating isolated pockets of oxygen depletion which can instigate localized corrosion through the establishment of differential aeration cells (Figure 4E) [Picioreanu and van Loosdrecht, 2002]. Roe et al. have demonstrated this effect [Roe et al., 1996]. They placed a μm -thick patch of agarose—a biogel with an oxygen diffusivity that is slightly lower than that of water—on to a low-carbon steel coupon, and observed localized corrosion beneath the abiotic film. Additionally, it has been proposed that enzymes embedded within the biofilm - such as hydrogenase - or other chemical properties of the biofilm may also enhance corrosion (Figure 4F) [Beech and Sunner, 2004]. The complexation of metal ions by functional groups, for example, is thought to enhance corrosion by altering the redox potential of ferrous ions within the biofilm [Beech and Sunner, 2004], though Roe et al. found no evidence to support this hypothesis [Roe et al., 1996].

In the presence of SRB, ferrous ions will also form iron sulfide corrosion products, the exact form and composition of which are difficult to predict. It is known that iron sulfides can transition from unstable, oxygen-reactive phases such as mackinowite to pyrrhotite and to stable pyrite FeS_2 [Rickard and Luther III, 2007], but in practice the conversion is spatially and temporally heterogeneous. Cwalina et al. have noted a difference in the structure of iron sulfides formed biotically versus abiotically, the former referred to as “amorphous precipitates” and the latter as “crystalline sediments” [Cwalina and Dzierzewicz, 1999]. Regardless, iron sulfides are cathodic toward iron, but this effect may be both protective and destructive [Ma et al., 2000, Stumper, 1923, Stott, 1993]. Much like the passive chromium oxide film found on stainless steel, a stable film of iron sulfide may protect the metal surface from further corrosion, but even a small disruption in this film can lead to a high rate of crevice or pitting corrosion due to the massive size disparity between the iron sulfide cathode and the exposed ferrous (iron or steel) anode. Of the various forms of iron sulfide, pyrite (cubic FeS_2) is known to be the most stable and also the most corrosive at any sites of film disruption [King et al., 1973a]; FeS_2 is also known to be susceptible to dissolution in the presence of certain bacteria such as *Acidithiobacillus ferrooxidans* [Rohwerder et al.,

2003].

The effect of a biofilm upon this passive film, however, is subject to much debate. Its presence may stabilize this passive film by acting as a physical binder and a protective coating. On the other hand, biofilm presence could potentially inhibit the formation and of such a film, or facilitate localized destabilization of such a coating (Figure 4G). Intriguingly, experiments by Lee et al. [Lee and Characklis, 1993] demonstrate both possibilities, differing only in the order of arrival of the biofilm versus the iron sulfide film. Less well-controlled, industrial environments such as oil pipelines include fluid flow and the continual formation and disruption of both iron sulfides and biofilms. In such contexts, the order of arrival for either film on the ferrous alloy, and thus the effect of the biofilm, remain ambiguous.

6 Summary and Outlook

Over decades of research from the perspectives of both corrosion and microbiology, significant progress has been to be made in understanding MIC. However, this mode of environmentally assisted corrosion remains a pervasive failure mechanism in a wide range of industrial contexts. We know more about the bacterial species and biofilms than ever before, but the complex interplay among species in extreme environments such as anaerobic fluid-metal interfaces has defied effective mitigation strategies. Renewed attention to the biofilm as a biotic coating that can potentially promote and mitigate corrosion of ferrous alloys in sour environments prompts several outstanding questions. In which ways do biotic and abiotic corrosion pits and rates differ? What combination of bacterial species and environmental factors promote MIC? How does fluid flow affect SRB biofilm stability, and how does this depend on the genetic diversity of that species? Answers to questions such as these will require new *in situ* assays of biotic corrosion, and will allow for improved predictions of environmental conditions susceptible to MIC. Together, these will facilitate cost-effective corrosion-risk monitoring. By approaching this challenging problem of MIC from both a materials engineering and a biological perspective, quantitative insights at the cell-material interface are poised to offer new understanding and mitigation strategies.

7 Acknowledgments

We gratefully acknowledge support from the MIT Energy Initiative (MITEI)-BP Energy Research Fund.

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List of Figures

1	MIC Damage on SAE 1020 Steel	20
2	SEM Images of Biofilm Growth	21
3	Nitrate Addition for Corrosion Control	22
4	Biofilm Mechanisms of Corrosion	23

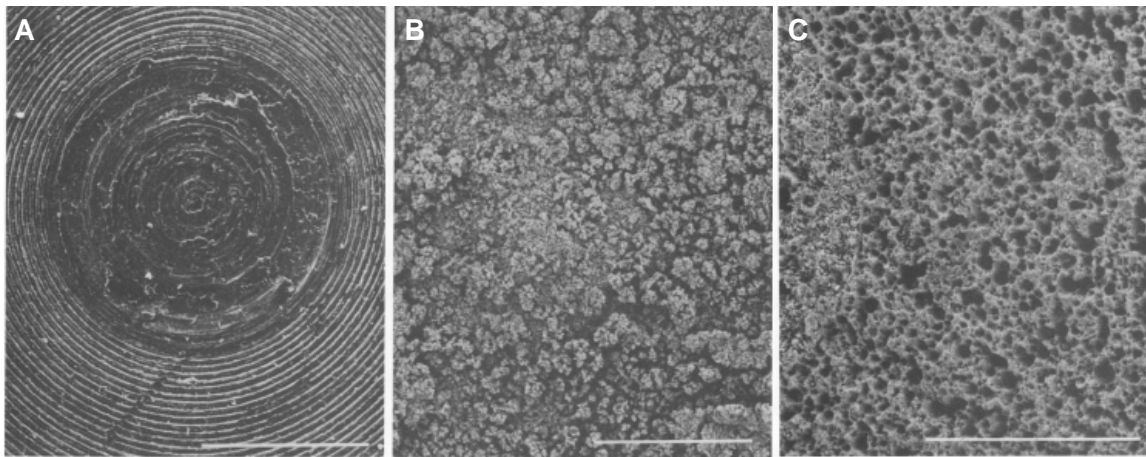


Figure 1: Scanning electron microscopy images which demonstrate the growth and pitting damage caused by sulfate-reducing bacteria growth on SAE 1020 steel studs. (A) Control stud showing concentric rings from lathe cutting; scale bar, 500 μm . (B) sulfate-reducing bacterial biofilm deposit after 64 days; scale bar, 500 μm . (C) Corroded stud after biofilm removal revealing severe pitting damage; scale bar, 500 μm [Bryant et al., 1991].

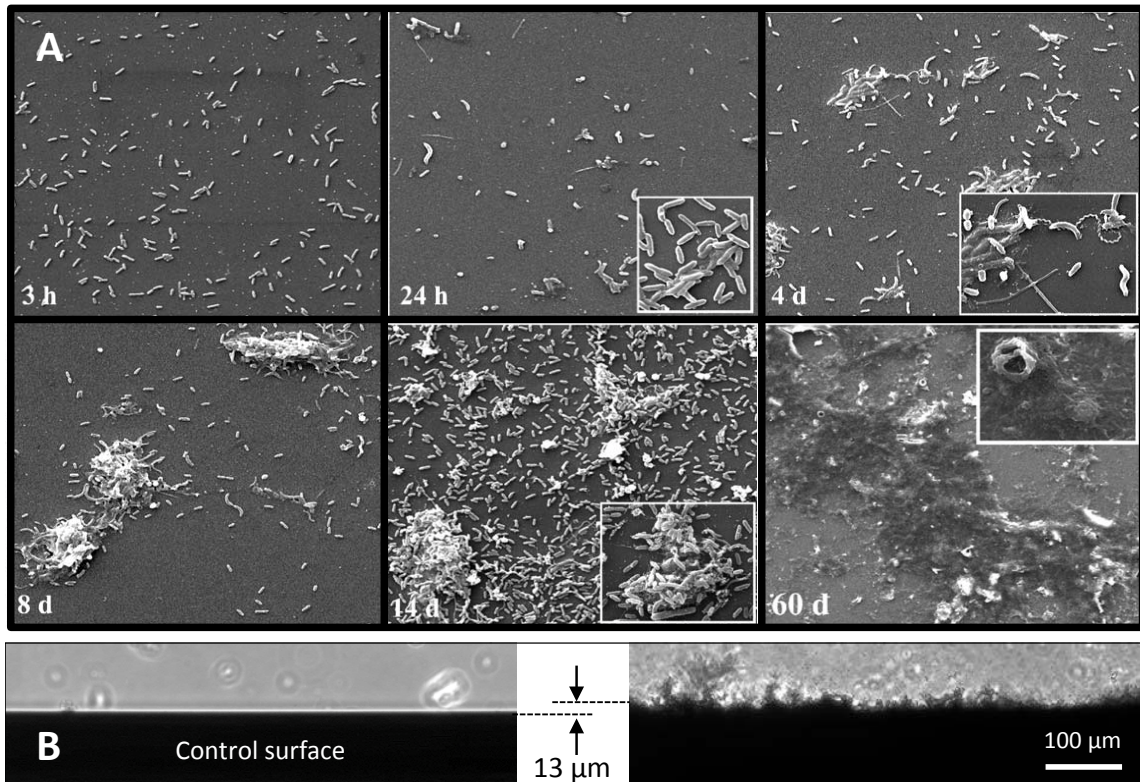


Figure 2: (A) Scanning electron microscopy images demonstrating the growth a biofilm over the course of 60 days. All images are at 1000x except for 60 days which is 200x. Inner boxes are at 4000x. [Fernandez et al., 2008]. (B) Original image demonstrating a cross section of the combined biofilm/corrosion product layer after 2 days of growth of *Desulfovibro vulgaris* on music spring quality steel (ASTM A228).

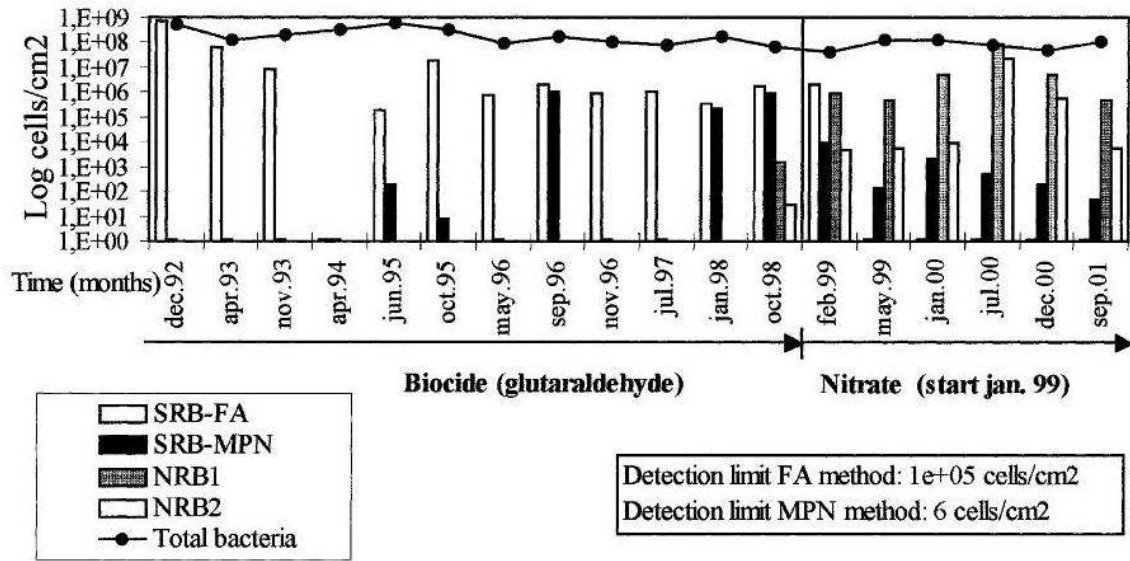


Figure 3: Measured data on the bacteria content of a floating unit at Veslefrikk, an oil production facility in the North Sea. Production began in 1989, utilizing a biocide treatment until January 1999 when it was stopped in favor of nitrate injection. A subsequent decrease in sulfate-reducing bacteria (SRB) and increase nitrate-reducing bacteria (NRB) numbers was observed. The population of SRB was characterized using a fluorescent antibody (FA) technique as well as the "most probable number" (MPN) method. Two types of NRB were measured using the MPN method: NRB1 which are facultative anaerobes, and NRB2 which are obligate anaerobes [Thorstenson et al., 2002].

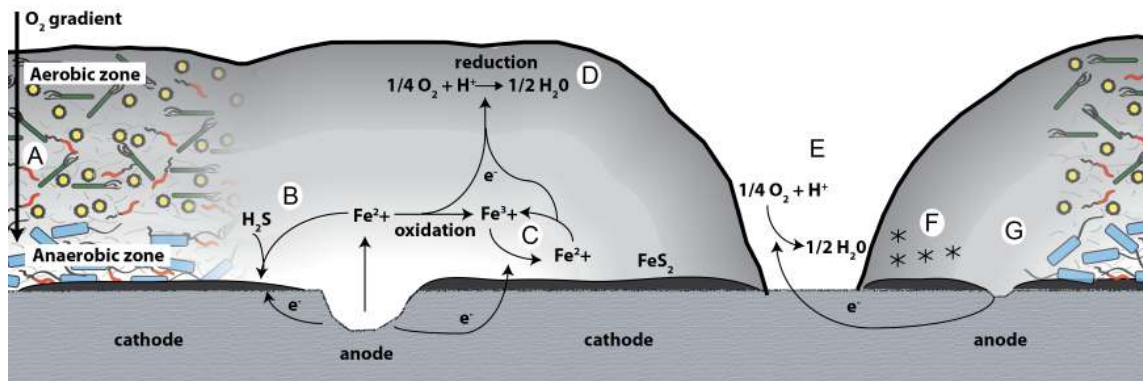


Figure 4: Mechanisms by which a biofilm can contribute to corrosion. (A) creation of anaerobic zones; (B) concentration of corrosive chemicals; (C) concentration of ferrous ions; (D) electron conduction away from surface; (E) creation of differential aeration zones; (F) binding of corrosion promoters; (G) disruption of passivating film.

List of Tables

1	Effect of Ferrous Ion Concentration on MIC	25
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Table 1: Relationship between ferrous ion concentration and corrosion rate, adapted from [King et al., 1973b].

Source	Ferrous ion concentration, <i>mM</i>	Average corrosion rate, <i>mg/dm²day</i>	Standard deviation
Booth & Wormwell [Booth and Wormwell, 1961]	0.025	4.04	1.75
Booth, Shinn & Wakerley [Booth et al., 1964]	0.125	3.0	-
Booth & Wormwell [Booth and Wormwell, 1961]	1.25	9.5	0.3
King, R.A. Ph.D Thesis, Univ. Manchester 1971	5.0	11.0	-
Bunker Report [Bunker, 1939]	10.7	17.5	1.5
Booth, Cooper & Wakerley [Booth et al., 1966]	12.75	20.87	3.39
King, Miller & Wakerley [King et al., 1973b]	20	21.2	2.47
King, Miller & Wakerley [King et al., 1973b]	38.2	24	-
Booth, Cooper & Cooper [Booth et al., 1967]	510	95	-