

BIOPROCESS ENGINEERING SYMPOSIUM — 1988

presented at

THE WINTER ANNUAL MEETING OF
THE AMERICAN SOCIETY OF MECHANICAL ENGINEERS
CHICAGO, ILLINOIS
NOVEMBER 27-DECEMBER 2, 1988

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THE AMERICAN SOCIETY OF MECHANICAL ENGINEERS
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MICROBIOLOGICAL CORROSION OF STAINLESS STEEL

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Abstract

The phenomenon of microbially influenced corrosion of stainless steel (Types 304, 304L, 316, 316L) is reviewed. The experimental techniques for the identification, isolation, development of bacteria cultures, the formation of biofilms, colonization and the nature of the environments created by the microorganisms are described. An electrochemical model is proposed to account for the corrosion of stainless steel in the presence of the microbes.

INTRODUCTION

In recent years, microbially influenced corrosion has been identified as a serious problem in the power generation industry, petrochemical industry, gas transmission lines and naval systems. The corrosion activity has been observed in sea water, untreated fresh water and even deionized water systems. Microbiological corrosion has occurred in service water systems fabricated from carbon steels, low alloy steels, stainless steels (Types 304, 304L, 316, 316L), copper-nickel alloys and high nickel alloys. However, systems using titanium appear to be resistant to microbial attack.

The purpose of this paper is to review the phenomenon of microbially influenced corrosion (MIC), including microbiological factors, electrochemical considerations, and effects on stainless steel.

MICROBIOLOGICAL FACTORS

An understanding of MIC begins with understanding the microbes that attach to the metal surfaces. The microbes which generate conditions that induce MIC are not those associated with biopharmaceutical applications. Rather they are a group of organisms which colonize the surface forming a biofilm in which they can generate organic acids, utilize oxygen, form polymers that chelate free metal ions, produce hydrogen sulfide and possibly even accept electrons from the metal surface. Understanding the MIC process begins with detecting the microbes on the surface. To show relationships with MIC, the microbes must both be found in the biofilm and have metabolic activity in that biofilm.

Detection of microbes: Microbes have the potential to influence corrosion by modifying the surface chemistry. One should suspect microbial involvement whenever the conditions allow the survival of microbes and some opportunity for their metabolic activity. One should strongly suspect MIC when there is highly selective damage or pitting, discoloration of the area, slimes associated with the failure, smells of microbial activities such as hydrogen sulfide, amines, mercaptans or development of obvious tubercles.

Bacteria involved in MIC are in surface attached biofilms. These biofilms consist of multiple organisms of many distinct physiologic types. The classical means to detect the presence of bacteria involves removing small portions of the surface biofilm and examining for microbes in the microscope, usually after staining with a dye like acridine orange that intensifies the contrast with the surrounding medium when exposed to ultraviolet light. The other classical means to find bacteria is to culture them on isolation (solid) media. The first technique, direct counting after staining requires that the bacteria be at concentrations $>10^5$ cells/ml. The second technique of growing the bacteria on solid medium requires that the bacteria be quantitatively recovered from the growth surface and be given a suitable growth environment in which they can form a detectable colony. The method we have developed and validated utilizes molecular probes as "signature" lipid biomarkers recovered from the membranes that surround each cell. These can be used to define the specific microbes that are present in the biofilm. If the system allows exposure to radioactive or mass isotope labeled precursors then the activity of these groups can be determined by finding the isotope incorporated into the specific signatures found in many of the bacteria. In many cases this can provide detection of the microbes under conditions in which the classical plate count is eliminated. Since the total community is examined in these procedures without the necessity of removing the microbes from surfaces, the microstructure of multi-species consortia is preserved.

Determination of the microbial biomass and community structure: The "signature biomarker" method involves the measurement of biochemical properties of the cells and their extracellular products. Those components generally distributed in all cellular life are utilized as measures of biomass. Components restricted to subsets of the microbial communities are utilized to define the community structure. The concept of "signatures" for subsets of the community based on the limited distribution of specific components has been validated by using antibiotics and cultural conditions to manipulate the community structure. The resulting changes agreed both morphologically and biochemically with the expected results (White et al., 1980). Other validation experiments involved isolation and analysis of specific organisms and finding them in appropriate mixtures, utilization of specific inhibitors and noting the response, and changes in the local environment such as light intensity. These validation experiments are summarized in reviews (White et al., 1986, White 1983, White 1988).

Phospholipids as molecular probe signatures: Phospholipids are found in the membranes of all cells. Under the conditions expected in natural communities, the bacteria contain a relatively constant proportion of their biomass as phospholipids (White et al., 1979a). Phospholipids are not found in storage lipids and have a relatively rapid turnover in some sediments, so the assay of these lipids gives a measure of the "viable" cellular biomass (White et al., 1979b).

Components of the phospholipids: The ester-linked fatty acids in the phospholipids (PLFA) are both the most sensitive and the most useful chemical measures of microbial biomass and community structure thus far developed (Bobbie and White 1980, Guckert et al., 1985). The specification of fatty acids that are ester-linked in the phospholipid fraction of the total lipid extract greatly increases the selectivity of this assay as most of the anthropogenic contaminants, as well as the endogenous storage lipids, are found in the neutral or glycolipid fractions of the lipids. By isolating the phospholipid fraction for fatty acid analysis, it proved possible to show bacteria in the sludge of crude oil tanks. The specificity and sensitivity of this assay has been greatly increased by the determination of the configuration and position of double bonds in monoenoic fatty acids (Nichols et al., 1986) and by the formation of electron capturing derivatives which, after separation by capillary GLC, can be detected by negative chemical ionization mass spectrometry at femtomolar sensitivities (Odham et al., 1985). This makes possible the detection of specific bacteria in the range of 10 to 100 organisms. Thus, analysis of the fatty acids can provide insight into the community structure of microbial consortia as well as an estimate of the biomass.

Use of the molecular probes in sulfate reducing bacteria: The sulfate-reducing bacteria, which are considered to be of particular importance in the microbial facilitation of corrosion, contain PLFA patterns which can be utilized to identify the lactate-utilizing Desulfovibrio, the acetate-utilizing Desulfobacter, and the propionate-utilizing Desulfobulbus (Edlund et al., 1985; Parkes and Taylor 1983; Taylor and Parkes 1983; Parkes and Calder 1985; Dowling et al., 1986). This allows differentiation between those utilizing lactate and propionate, or those using acetate and higher fatty acids. PLFA biomarkers for sulfate-reducing bacteria and indications of sulfate-reducing activity were readily detected in anaerobic fermenters supplemented with sulfate. Using this technology we were able to initiate studies of MIC consortia by incubating stainless steel coupons in a galvanic corrosion cell in aerobic seawater. The results indicated that it was possible to set up a consortium of a facultative Vibrio and the anaerobe Desulfobacter that was much more corrosive than either monoculture. Furthermore, it was possible to create the consortia in aerated sea water even though the Desulfobacter is a strict anaerobe. We were able to establish that a consortium can create anaerobic microniches in aerated systems that facilitate MIC.

Bacterial activity: Not only must specific bacterial consortia be present, but there must be metabolic activity for MIC. Estimates of microbial activity can be derived from the nutritional status as reflected in the lipid analysis. The nutritional status of certain bacteria in biofilms can be related to shifts in the metabolic activities that could potentiate corrosion processes. The nutritional status of microbial consortia can be estimated by monitoring the proportions of specific endogenous storage compound relative to the cellular biomass. Certain bacteria form the endogenous lipid poly beta-hydroxyalkanoate (PHA) under conditions when the organisms can accumulate carbon but have insufficient total nutrients to allow growth with cell division.

A second measure of community nutritional status is the formation of extracellular polysaccharide glycocalyx. Uronic acid-containing glycocalyx forms maximally in the marine Pseudomonas atlantica under conditions of nutritional stress (Uhlir and White, 1983). Uncontaminated subsurface aquifer sediments contain microbiota with very high levels of extracellular polymers form on the surfaces of metals exposed to rapidly flowing seawater and may be responsible for inducing reversible acceleration of corrosion (Nivens et al., 1986).

Starvation induces the formation of minicells in some marine bacteria. There is a loss of cell components including the membrane lipids, but there is a marked increase in the proportion of monoenoic PLFA with the double bond in the trans configuration (Guckert et al., 1986) and is formed from 14-C acetate (Guckert et al., 1987).

Since the signature methods involve the isolation of specific components, the rates of incorporation of labeled precursors can be utilized to detect the metabolic rates of specific components by measuring the increase in non-radioactive 13-C labeled PLFA from a specific group of bacteria using the extraordinary resolution and sensitivity of gas chromatography/mass spectrometry (GC/MS).

Non-destructive analysis: The GC/MS methods based on quantitative analysis of components of the microbiota and its extracellular polymers has shown correlations to electrochemical analysis of corrosion, but it offers no on-line independent methods for correlation. The increases in sensitivity of infrared spectrometry with the application of Fourier transforming infrared spectrometry (FT/IR) provides a technique for analysis of living biofilms. Attenuated total reflectance (ATR)-FT/IR allows examination of living biofilms. ATR-FT/IR detects vibration-rotation interactions of biofilms between about 0.5 and 1.5 μm outside the surface of the germanium crystal used in the ATR cell. With this system it has proven possible to show that the carbohydrate rich initial fouling polymer coats the germanium surface exposed to sterile seawater in about 13 hours (Nichols et al 1985). With this system it has proven possible to monitor the development of biofilms by flowing bacterial cells and nutritive media through the

ATR cell. Exopolymer polysaccharide was formed more rapidly than the proteins of the bacterial cell. Protein formation appeared to be more sensitive to growth conditions than polymer formation. The system is being utilized to monitor the penetration of biocides into living biofilms.

If the biofilm is first freeze dried then the resolution of the FT/IR analysis can be increased considerably although the ability to monitor living biofilms is sacrificed. The technique of diffuse reflectance infrared Fourier transforming (DRIFT) can be used to detect two physiological changes identified with compromised microbiological nutrition. Two physiological changes have been identified as markers for microbial nutritional status: The formation of PHA and the uronic acid-containing exopolysaccharide glycocalyx which are responses to nutritional stress in bacteria. Both polymers can be detected with the FT/IR. Using standard addition experiments, shifts in the amounts of PHA and glycocalyx were demonstrated in biofilms (Henson et al., 1985).

The DRIFT can be utilized with an infrared microscope with the freeze dried biofilm on a corrosion coupon. The microscope allows mapping the ratios between bacteria, PHA, and exopolysaccharide polymers to a resolution of 20 μ m in diameter. The exposure of 4 cm diameter polished 316 stainless steel autogenous weldments in the continuous shear gradient of the sterilizable Fowler cell adhesion module to growing cultures of the marine bacteria *Pseudomonas atlantica* resulted in adhesion in a gradient. More bacteria attached to the welded area than the base metal, and the bacteria attached to the weldment in the highest shear showed a higher exopolymer to cell protein ratio than the cells on the base metal or those attaching in lower shear environments measured by DRIFT. The PLFA patterns were significantly different in the cells attaching in the high shear area than those that did not attach and those that attached to the areas of low shear.

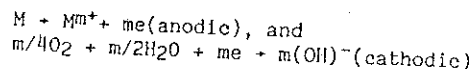
Stainless steel weldments showed higher densities of bacteria attached to the weld than the base metal (estimated by DRIFT and by PLFA analysis of recovered biofilms by GC/MS). Prior to these estimations of the localizations of microbes on the weldment as apposed to the base metal the coupons had been used as the working electrodes in electrochemical impedance spectrometric (EIS) measurements. EIS analysis showed corrosion that gave evidence of localization in the low frequency response. Open cell potential monitoring showed a marked change. These changes correlated with the appearance of pitting corrosion at the boundary of the fused zone and the heat effected zone. Examination of the area with electron spectroscopy for chemical analysis (ESCA) showed much higher concentrations of Ni and Cr associated with the area of pitting than in other areas. The electrochemical analysis, the microscopic examination, and ESCA all showed evidence of pitting corrosion with specific loss of the iron in the fused zone. Examination of the coupon showed that this zone contained a distinct microbial consortia different from the microbes on the base metal.

With this information on the microbiological factors, the electrochemical behavior of materials in the presence of the microbes is examined in the following section.

ELECTROCHEMICAL CONSIDERATIONS

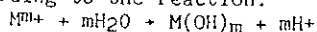
Relative to the microbially-influenced corrosion susceptibility of metals and alloys in fresh-water systems, a working hypothesis is proposed. Overall, it is believed that the same electrochemical reactions are involved with MIC pitting corrosion as with classical deposit/crevice corrosion and subsequent pit initiation. The major difference is that several critical steps in the overall mechanism are accelerated or enhanced by microbe attachment, biofilm formation, and the metabolic activities of microorganisms. Thus, the classical electrochemical mechanisms are operative, but through synergistic microorganism effects, produce more severe environments at the metal/solution interface in shorter time intervals than would occur under standard deposit/crevice corrosion conditions. Detailed steps in the hypothesis, and supporting laboratory data, will be given in the following sections.

Figure 1 summarizes the critical elements of the working hypothesis on microbially-influenced pitting corrosion. Electrochemical events in time sequence are shown. First, events associated with the classical deposit/crevice corrosion mechanism will be summarized. In this description, the prototype metal will be a stainless steel which is naturally passivated with a chromium-oxide-rich passive film in the bulk aqueous solution. The process starts with the formation of a crevice geometry, which could be formed in a number of ways including valleys and/or overlaps in a rough surface, the interface at a mechanical connection, etc.; but in this description the crevice geometry is assumed to be formed by an inert deposit (e.g. dirt, oxide particle swept in from another location, etc.). The point in approaching the hypothesis in this direction is to later compare the effects of this inert deposit with those of a living, microorganism-type "deposit". Under the inert deposit in a near-neutral, aerated, bulk aqueous solution, initially the following oxidation and reduction reactions occur at the same electron-producing and electron-consuming rates at local anodic and cathodic sites:



However, within the crevice geometry, the cathodic reaction involving reduction of dissolved oxygen soon slows or stops due to diffusion limitations associated with movement of oxygen into the tight, stagnant, crevice area. Thus, the production of $(OH)^-$ anions slows or stops. However, the production of M^{n+} cations by the anodic reaction continues, provided other anions can diffuse to the crevice region to maintain electroneutrality of the local crevice solution. The chloride ion has one of the fastest diffusion rates, and if the bulk solution

contains chloride ions, they will diffuse to the crevice solution to balance the charge of the M^{m+} cations being produced, resulting in a much higher chloride ion concentration, $[Cl^-]$, within the crevice solution as compared to the bulk solution. An additional reaction in the crevice solution occurs. As the metal-ion concentration increases, it can reach a solubility limit (dependent on pH and $[Cl^-]$). If the solubility is exceeded, water hydrolysis occurs according to the reaction:



Due to this hydrolysis reaction, acidification of the local crevice solution occurs. In summary, the classical deposit/crevice corrosion mechanism, as indicated in Figure 1, involves: (1) formation of the crevice geometry, (2) O_2 depletion, and (3) chloride concentration and acidification of the local crevice solution. If the crevice solution becomes sufficiently severe, i.e. in terms of low pH and high $[Cl^-]$, the protective passive film on the stainless steel will be broken down, causing the initiation and autocatalytic propagation of pitting corrosion. Whether or not the passive film is broken down depends not only on the severity of the local crevice solution but also on the inherent resistance of the passive film to dissolution under conditions of low pH and high $[Cl^-]$.

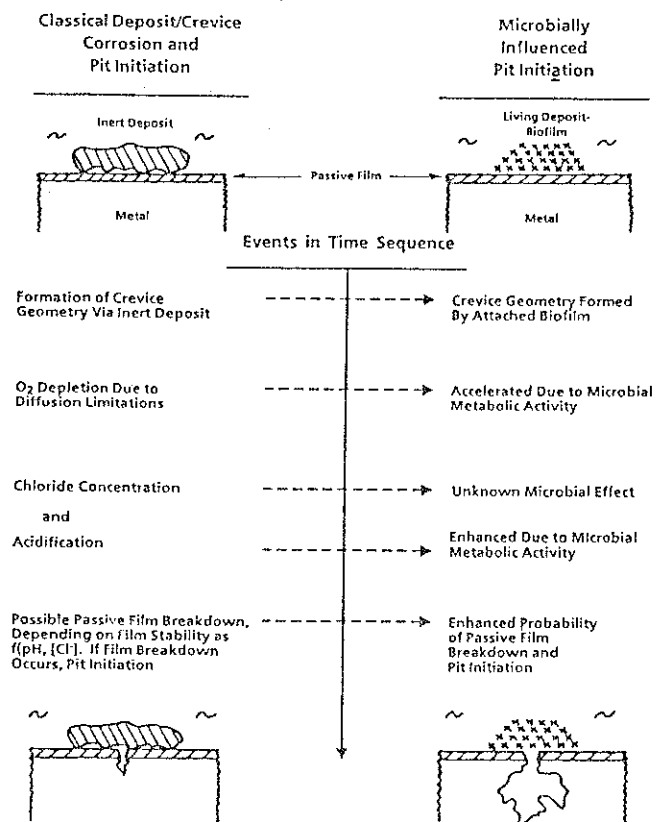


Figure 1. Working hypothesis, microbial influence on pit initiation.

Now the hypothesized influence of microorganisms on the overall process will be introduced. First, as indicated in Figure 1, it is believed that through non-uniform formation of biofilms at the surfaces, the microorganisms by their natural attachment and agglomeration actions produce crevice-type geometries, thus substantially increasing the probability of the first step in the previously-described classical mechanism. Next, aerobic microbes metabolically consume dissolved oxygen, thus accelerating the oxygen-depletion process relative to the classical mechanism. Third, many microorganisms, both aerobic and anaerobic, produce acids through metabolic processes, thus accelerating acidification of the local crevice solution. (Other metabolic effects which accelerate electrochemical reactions are also possible: e.g. chelation of metal ions produced by the corrosion process, and as a consequence, increased anodic dissolution rates; or production of highly oxidizing species such as ferric ions, which would cause an increase in the corrosion potential, an increase in the anodic dissolution rate, and possibly the initiation of pitting corrosion.) The overall effects of these microbial enhancements would be either to accelerate the onset of pitting corrosion because of a more severe local crevice environment, or to "cause" pitting corrosion (i.e., for this latter situation, one would consider that the local crevice environment under classical conditions was insufficiently severe to break down the passive film and induce pitting corrosion).

Several types of preliminary laboratory results are presented in support of the above working hypothesis. The first set of data involved measurement of open-circuit corrosion potential, E_{corr} , as a function of time for several structural alloys in growth media with and without the presence of certain microorganisms. The alloys included types 304L and 316L stainless steel, admiralty brass, and 90-10 copper-nickel alloy. The growth medium was Hutner's medium (Manual of Methods for General Microbiology, American Society for Microbiology). The microorganisms consisted of either a gram negative, heterotrophic, facultatively anaerobic, acid producing bacteria (A6F) extracted from tubercles associated with MIC failures in untreated-water systems at a certain utility plant site, or a consortium of five bacteria extracted from untreated water known to be causing MIC problems at a different utility site. The intent in these studies was to determine if changes in E_{corr} , as a consequence of pit initiation and propagation, were sufficiently large to be easily measured. Results for 304L are given in Figure 2.

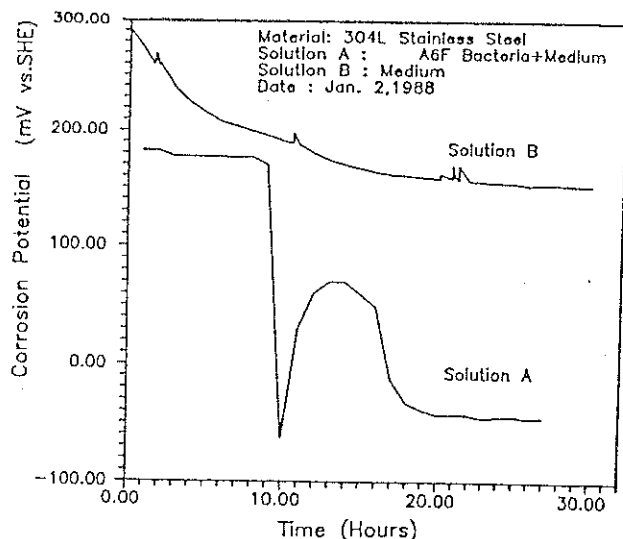


Figure 2. Open-circuit corrosion potential in solutions without bacteria (B) and with bacteria (A).

As is evident, large differences in E_{corr} were produced, and subsequent surface analyses showed that corrosion pits were propagating in the medium plus concentrated-bacteria solution but that corrosion pits were propagating in the medium plus concentrated-bacteria solution but were not initiated in the medium only. For the present discussion, however, a more applicable type of result was revealed. In these experiments, it was also decided to continuously monitor the open-circuit potential of a platinum electrode. A common result was observed for both types of microbial solutions employed: the platinum potential underwent a sharp, major reduction after an incubation period of time. Typical results are shown in Figure 3 for the consortium of five microorganisms.

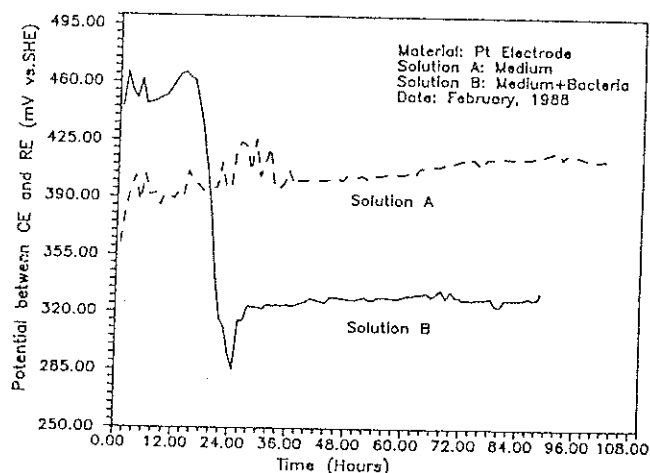


Figure 3. Open-circuit potential of platinum in solutions without bacteria (A) and with bacteria (B).

In no case did the platinum potential undergo such a reduction in the growth medium, i.e. the control solution, which did not contain the bacteria. Filtered compressed air was

continuously bubbled through both medium and medium-plus-bacteria solutions; thus, the bulk solutions were aerated. Nevertheless, the sharp reduction in platinum potential in the bacterial solutions clearly indicated a major reduction in dissolved oxygen at the platinum/solution interface. This effect is believed to be due to biofilm formation, with the development of crevice-type geometries, and also due to metabolic processes within the biofilm which consume oxygen. Therefore, this experimental evidence is taken as supportive of certain elements in the working hypotheses involving microbially influenced pitting corrosion--that microorganisms accelerate the development of crevice-type geometries and accelerate oxygen depletion within the local crevice solutions. Also as part of these preliminary experiments, the bulk solution pH was measured before and after exposure. Whereas the pH of the medium changed very little during the exposure time, the pH of the medium-plus-bacteria solutions changed from near-neutral values to bulk-solution values of pH=4 in one week. It is reasonable to assume that the pH values were even more acidic at the biofilm/metal interface. This evidence, too, is taken as supportive of the working hypothesis--that microorganisms accelerate acidification of the local crevice solution.

Given that the microorganisms produce environmental conditions which accelerate the electrochemical process, the effect of MIC on stainless steel is presented in the last section of this paper.

EFFECT ON STAINLESS STEEL

Types 304, 304L, 316 and 316L

MIC of austenitic stainless steels, has been reported in many of the service water lines in the utility industry (Licina and Giannuzzi, 1986). The MIC characteristics and the metallurgical factors are reviewed in this section.

A recent Electric Power Research Institute (EPRI) source book prepared by Licina (1988) covers five case histories of MIC in austenitic stainless steel pipes. These involved Types 304 and 304L stainless steel base metal and 316L weld metal. Pitting attack occurred in base metal, heat-affected zones and weld metal. In some cases there was preferential attack of the delta ferrite of the weld metal while in other welds the austenite was selectively dissolved. Borenstein and Lindsay (1987) reported results of MIC of Types 304, 304L, and 316L stainless steel piping. Pitting corrosion occurred in class ER-308 weld metal, heat-affected zone and base metal. There was evidence of selective dissolution of the delta ferrite in the 308 weld metal. Licina (1988), in a review article reported that MIC in the austenitic stainless steels is characterized by pitting. Attack occurs predominately in weld metal, heat-affected zones and to a lesser extent in base metal. In the two phase (austenite and delta ferrite) weld metal, there has been preferential attack on each of these phases.

No explanations have been offered on the preferential dissolution of the austenite and delta ferrite phases in the weld metal. Metallurgical differences between the two phases may contribute to the selective dissolution. Austenite chemistry is high in nickel content while the delta ferrite is enriched in chromium. In the as-welded condition non-equilibrium conditions are produced that result in microsegregation of the alloying elements in both phases. These chemical variations may result in localized electrochemical cells during MIC.

The weld metal also has high tensile residual stresses present in the as-welded condition. These stresses provide a source of high energy levels that may contribute to the kinetics of the MIC process. In the heat-affected zones of weldments of Types 304 and 316 stainless steels, a sensitized microstructure is produced. Chromium rich carbides (Cr₂₃C₆) precipitate at the austenite grain boundaries and make the boundaries susceptible to intergranular attack. Depending on the severity of sensitization, the localized chromium content at the grain boundaries may be at levels that no longer provide passivation. This could contribute to the MIC in the heat-affected zones. The sporadic appearance of MIC in the base metal may be in part related to microsegregation of alloying elements and residual elements that occur in the mill annealed condition and in part to severe localized chemistries from the presence of microbes that exist in creviced areas.

SUMMARY

Microbially influenced corrosion occurs in austenitic stainless steels, (Types 304, 304L, 316, and 316L). MIC is manifested by pitting reactions primarily in the weld metal, heat-affected zones and to a lesser extent in the base material. The two phase weld metal composed of austenite and delta ferrite has shown preferential attack of the austenite in some cases and selective dissolution of the ferrite in other failures. The phenomenon of MIC consists of a number of phases that include: a source of water that contains microbes, the attachment of the microbes through the formation of biofilms, the establishment of microbial consortia or colonization and production of an environment to create and sustain corrosion. A model based on classical electrochemical concepts is proposed to describe the phenomenon. Application of these principles does not fully explain the MIC at various locations within the stainless steel, specifically, weld metal, heat-affected zones, base metal and the selective dissolution of austenite and delta ferrite in the weld metal.

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